Asian Longhorned Beetle

Proposed FY 2002 Research & Technology

Table of Contents

Asian Longhorned Beetle
Research and Technology Development

Introduction
The Asian longhorned beetle (ALB) eradication program has four basic components: survey, regulatory, control, and program management.

A. *Survey* determines the size and scope of the outbreak and where regulatory and control activities are required.
B. The *regulatory* component prevents the artificial or human assisted movement or spread of the pest out of the infested areas while eradication is completed. At ports of entry, it also prevents subsequent reintroduction from foreign sources.
C. *Control* involves a combination of biological, cultural and chemical methods and treatments to eliminate the pest.
D. To effectively direct and manage resources, *program management* depends upon the development of operational guidelines, protocols, and strategies based on sound science, and data collection and management systems to assist decision-making, track progress and measure performance.

Little was known about the ALB when it was first introduced into the United States in 1996. Scientists and technical experts have since provided the necessary information and developed the tools and methods to implement an eradication program. Although the program is progressing in its goal to eradicate ALB, there are many areas within each component where significant improvement is possible. Continued support from scientists and technical experts is needed to reduce costs, improve efficiency and ensure successful eradication.

A. **SURVEY**

Program

Survey is by far the most expensive component of the program and arguably the most critical. It will extend throughout the life of the program. Survey activities determine where other activities are to be conducted, track the effectiveness of those activities, and verify and validate eradication. Currently, all survey is conducted through visual observation of individual host trees. Inspection by climbing the tree or utilizing bucket trucks is much more effective than from the ground but also much more expensive. Multiple inspections of each tree over several years will be required using current technology and methods.

Goal: Develop more effective and/or cost efficient ALB survey systems.

Approach: Survey can be improved through: improving visual survey methods and technology based on understanding ALB dispersal and behavior and host location mechanisms; developing a trapping system; utilizing novel approaches such as acoustic detection or remote sensing technologies; and designing and optimizing survey systems for detecting and delimiting ALB populations.

OBJECTIVE AND JUSTIFICATION:
Improve current visual survey methods and technology based on understanding of dispersal and other behaviors, including host location mechanisms.

The effectiveness and efficiency of visual surveys for ALB can be enhanced by maximizing the chances of finding the insect or its damage when inspecting infested trees and by optimizing the allocation of survey resources within and beyond the program areas. Decisions on areas to be surveyed, and on the intensity of survey within those areas, need to incorporate biological information such as distance beetles will move from sites of known infestation, and what types of trees beetles prefer to attack and utilize. This objective relies heavily on understanding the fundamental biology of the pest, including dispersal and movement behavior; and host preference, condition, and availability. Alternative sampling strategies (e.g., tree risk ratings, sentinel trees) should also be considered.

Understanding dispersal potential of ALB requires studies of both population dispersal potential and individual movement of adult ALB. These studies include determining flight propensity (probability of initiating flight) and flight capacity (ability and probability of distance flight). These studies also require investigations of dispersal and movement along a temporal continuum, including instantaneous, daily and seasonal movement. Finally, the key factors that influence flight propensity and capacity must also be evaluated. These include ALB population density, beetle sex, size and age, host tree characteristics, tree health, landscape characteristics, as well as environmental variables (*e.g.,* temperature, humidity, wind). Quantifying these factors and their influence will facilitate the prediction of dispersal in urban landscapes currently infested by ALB in the U.S., as well as in other landscapes at risk in North America. This proactive approach will form the basis for development of adaptive management strategies for this and other invasive species. Understanding the host location mechanisms of ALB requires investigations of the cues by which ALB searches for, orients to, and lands on host trees. Collectively, this is referred to as in-flight host selection. The key factors that influence host selection must also be evaluated. These include ALB sex and age, host tree (characteristics and health), landscape characteristics, as well as environmental variables. Both laboratory and field studies will be conducted. Field studies will be conducted in both urban and forest settings to determine if urban background (visual) noise influences the use of visual and/or olfactory signals.

Understanding ALB colonization behavior, including mating, oviposition and feeding, requires investigations of the attack pattern and resulting within-tree ALB distribution, and the emergence pattern and the associated resident population density (all of which have spatial and temporal qualities). Furthermore, understanding these behaviors requires identification of the factors that govern them, so as to ensure their exploitation for eradication of ALB.

Work plans:
ARS: ARS 02.03; ARS 02.08; ARS 02.11
Forest Service: FS 02.13
APHIS: APHIS 02.10; APHIS 02.11; APHIS 02.15; APHIS 02.16

OBJECTIVE and JUSTIFICATION:
***Develop a trapping system to aid in the detection and delimitation of ALB*.**

A trap is a device that captures organisms and holds them in a location where the trapper can find, count, and/or otherwise process them. Although some traps are passive (the target organism must "stumble into them"), trapping systems for insects typically include one or more means of attracting the target species to the trap. These means may be visual (shape, color, light-emitting) or chemical (odors such as pheromones or volatiles from host plants). A good attractant can greatly increase the effectiveness of a trapping system to the point where the system becomes a sensitive and cost-effective tool for detecting whether or not a population of the target insect is present. In addition, the design of the trap itself, including the means by which it actually captures the insect, greatly influences the effectiveness of the system. Answers to fundamental biological questions on the pest's behavior, dispersal, host preference, and physiology are necessary to design effective traps as well as protocols for deploying them. Attractant-based trapping systems have proven themselves repeatedly as highly sensitive tools of choice in detection surveys for such regulatory pests as tephritid fruit flies, gypsy moth, bark beetles, and boll weevil. An effective trapping system would simplify ALB survey operations, improve the sensitivity of the survey, and reduce costs dramatically. Unlike visual surveys, traps monitor an area continually.

A GC-EAD (coupled gas chromatograph and electroantennogram detector) has been used to find over 20 components of ALB odor and ALB host-plant odor that could be sensed by the adult beetles. Over half of those components (individual chemicals) were subsequently identified chemically by mass spectroscopy (identification of the remainder is ongoing). Those chemicals are being tested in laboratory assays and in field trials in China (along with prototype traps) to see if one or combination of several may provide useful attractants for trapping or baiting. Understanding how beetles respond to visual cues (color, shapes), when and how they move (fly, walk), and where they spend their time is critical to developing and optimally deploying traps. Because any field studies in this area require observations on large numbers of beetles in natural settings, they must be conducted in China.

Work plans:
ARS: ARS 02.01
Forest Service: FS 02.04; FS 02.05; FS 02.10, FS 02.13, FS 02.14; FS 02.17;
FS 02.19; FS 02.20
APHIS: APHIS 02.06; APHIS 02.08; APHIS 02.09

OBJECTIVE AND JUSTIFICATION:
Develop novel approaches for survey such as acoustic detection or remote sensing technologies.

Acoustic detection has great potential for both the eradication project, and as part of exclusion activities at ports of entry. For eradication, a primary use would be to place acoustic detectors in close proximity to infested trees to discover additional infested trees that were not detected through visual inspection. Removal of these additional infested trees would suppress the pest population and expedite eradication.

Acoustic signals are generated as ALB larvae feed and tunnel in wood. The specific acoustic signals created by a wood-boring insect are likely to be related to its size, the shape of it mouthparts, its feeding behavior, the configuration of the gallery or tunnel that it constructs, the species of tree infested, and the type of infested material, i.e., a live tree or lumber. Therefore it is likely that a unique acoustic signature for ALB-infested trees or wood exists. The ultimate objective of this work is to determine unique ``Acoustical Signal Descriptors'' that indicate the presence of ALB and to develop a field-portable system using these descriptors that can detect ALB in live trees and wood. It is anticipated that each tree species will have a somewhat unique frequency range and attenuation factors given its particular cell structure and moisture content. Using infested trees and wood, scientists will determine the amount of energy generated by a single feeding larva.

Scientists will also obtain hi-fidelity data in the audio range for actively feeding ALB larvae in trees and wood. They anticipate that the frequency range and energy level will change as the larva grows, given that the size of the cavity holding each larva will alter the resonance frequency to some degree. At some point in the larval growth cycle, the signal frequency may drop from the ultrasonic range into the audio range; then, microphones with parabolic reflectors can be used to measure sounds from the trees. Scientists will also record hi-fidelity data in the audio range (20 Hz to 20 kHz) of ALB adults as they chew their way out of the trees and as they chew pits through the bark in which they then lay eggs. In addition, we will obtain ultrasonic and audio frequency range data of a few other common wood boring beetles. This information will be necessary to determine the uniqueness of the acoustic signal descriptors of ALB when compared with other wood-boring beetles.

Once the recordings have been made, extensive analyses of the ultrasonic and acoustic data will be conducted to develop algorithms for identifying the presence of ALB. Scientists will then design, produce, and test various prototype instruments that embody the ALB-identification algorithms. Finally, they will develop a hand-held instrument and verify its ability to identify the presence of ALB under field conditions. Equipment innovations include development of a lightweight acoustic detection kit for ALB larvae with signal interpretation.

Obtaining access to host trees is a significant problem in New York especially in Manhattan. A great amount of resources is spent trying to contact property owners to identify properties with hosts and obtain permission to survey or treat. Inaccessible terraces and backyard areas could act as reservoirs for the pest delaying eradication.

Remote sensing may be used to identify properties of host trees in inaccessible areas. Remote sensing will not likely detect individual infested trees but may be a valuable planning tool to estimate workload, and support survey and treatment activities. Moreover, it may prove to be an effective means of rapidly screening large areas for “pockets” of trees that show stress that may be indicative of ALB infestation. This capability could greatly improve the efficiency of visual surveys for “satellite” populations, thus increasing the chances of locating and intervening at such infestations before they become unwieldy and/or spread to other sites. In initial research in this area, the potential of remote sensing technology for likely sites of ALB infestations will be investigated by evaluating images from available libraries for relationships between spectral plant stress indicators and sites of previous ALB finds.

Work plans:
ARS: ARS 02.04; ARS 02.05; ARS 02.06; ARS 02.07
Forest Service: FS 02.10; FS 02.04; FS 02.17
APHIS: APHIS 02.19

OBJECTIVE AND JUSTIFICATION:
Design and optimize survey systems for detecting and delimiting ALB populations.

The tools that are used to detect insect populations (e.g., traps, visual surveys methods, acoustical devices) form only one part of a survey system. An equally important component is knowledge of how to use those tools effectively and efficiently to produce the most sensitive survey system possible within limits imposed by the availability of resources or other constraints. Detection of an insect infestation is a probabilistic event, and the probability of detection is influenced by such factors as the sensitivity of the detection tool, the size of the infestation, the distribution of the insects (and the sampling effort) in time and space, and the overall intensity of the sampling effort.

Available information on the insects’ behavior, movement, and seasonality will be analyzed and incorporated into spread and detection models to estimate optimal strategies for survey protocols. During this process, gaps in available data have been and will continue to be identified; these gaps will be filled both by studying available program data on occurrences and location of beetles and beetle damage and by conducting field studies on beetle dispersal and distribution in China.

In addition, strategies for quality control and quality assurance for ALB surveys will be developed and implemented in operational programs. Efficiency of visual and other survey methods will be assessed experimentally, and results will be incorporated into estimates of efficiency of detection efforts. Methods of producing “false positive” survey results (without compromising program integrity) are under development and will be used both in efforts to assess survey sensitivity and as a means to assure a high level of performance from survey crews.

Work plans:
ARS: none
Forest Service FS 02.05, FS 02.17, FS 02.19, FS 02.20, FS 02.13
APHIS: APHIS 02.10; APHIS 02.11; APHIS 02.15; APHIS 02.16

B. REGULATORY & EXCLUSION

Program

Stopping additional ALB infestations from establishing is a critical component of our efforts to return the United States to an *Anoplophora*-free country. To that end, APHIS now requires (since December 17, 1998) that all solid wood packing materials (SWPM) from China and Hong Kong be treated to eliminate wood-boring insects (7CFR 319.40). This requires the availability of effective treatments that are compatible with Chinese SWP manufacturing practices. In addition, APHIS inspectors need effective methods for detecting the presence of ALB in imported wood items and for confirming that SWPM were treated in accordance with existing regulations.

Goal: Provide technology to minimize the risk of introducing ALB into uninfested areas.

Approach: Before the discovery of ALB in the U.S., there was very little information on regulatory treatments specific to ALB. Currently approved treatments will be reviewed to confirm their effectiveness against ALB, and new, simple, and inexpensive methods of disinfesting SWPM are being developed for transfer to Chinese exporters. In addition, acoustical and chemical methods are being investigated as possible tools to help APHIS inspectors detect ALB efficiently in imported wood packing and commodities.

OBJECTIVE AND JUSTIFICATION:
Systematics and detection of wood-boring insects in imported materials (using chemicals, ultrasound, x-ray, acoustics and IR technologies).

The ability to interdict insects in SWPM is critical to stopping ALB-infested materials from entering the U.S. Improved systematics (including expert identification systems for on-site use) and improved detection systems are needed. Currently, ALB and associated damage are detected only through visual inspection of cargo, which often requires complete de-vanning at the point of inspection. Even when employed, this is prohibitively expensive and time-consuming in the vast majority of cases, and it is not completely effective because the insects can reside in internal portions of large SWPM items such as cable spools.

ARS and FS scientists are investigating the use of acoustic devices for detecting chewing insects in wood. In these studies, the approach has shown great promise for detecting the presence of ALB larvae in standing trees, and could readily be adapted to SWPM and imported commodities such as bonsai and penjing plants. The two agencies (and APHIS; see NOTE 1) are investigating the possible value of different types of acoustic sensors in hopes of finding at least one that is effective and perhaps several that may be best suited for specific applications. Other high-tech approaches to this problem are also being investigated; for example, FS is looking for a unique chemical profile to odors associated with ALB larvae; such odors could be used to detect the presence of ALB in shipping containers. In this research, scientists will identify unique volatile organic chemicals (VOC's) associated with the presence of the ALB in live trees and wood; and develop a portable ALB detection systems based on ALB-specific VOC's. Scientists will employ active air sampling onto solid sorbents followed by analysis using thermal desorption gas chromatography/mass spectrometry to search for unique marker chemicals and chemical signatures. Sampling onto ``Triple Sorbent Traps'' followed by traditional capillary column gas chromatography and electron impact mass spectrometry has proven effective for the identification of terpenes and other VOC's released by plants. Airborne concentrations of VOC's in the low part-per-billion range (generally picogram quantities) are detectable.

Because ALB infests several species of trees, it will be necessary to sample different species of infested and uninfested trees so that statistically significant signatures can be identified. Similarly, we will sample VOC's from infested and uninfested wood from different tree species.

Work Plans:
ARS: ARS 02.02 (related: ARS 02.01; ARS 02.04; ARS 02.05; ARS 02.06; ARS 02.07)
FS: FS 02.04; FS 02.10
APHIS: none (see NOTE 1)

> *NOTE 1: APHIS-PPQ-CPHST has initiated additional work plans in this area in response to the ALB situation. These include studies on (1) acoustic detection of insects in imported packing materials and commodities in general, and (2) the use of non-contact ultrasound to detect internal damage and organisms in wood packing materials and other commodities. Because applicability of these projects extends beyond the scope of ALB eradication programs, they are being funded through alternate mechanisms (specifically, AQI user fees).*

OBJECTIVE AND JUSTIFICATION:
Provide effective regulatory treatments for eliminating Anoplophora in imported materials.

Current regulations state that SWPM from China (including Hong Kong) must be (1) fumigated with methyl bromide, (2) treated with wood preservatives, or (3) kiln dried before it will be allowed to enter the United States. APHIS-PPQ-CPHST has been working to reconfirm the effectiveness of current fumigation schedules. This is a critical need, as was demonstrated by the withdrawal of an existing schedule for phosphine fumigation after it was found to be inadequate for killing ALB in wood packing. In addition, given potential problems with future availability of methyl bromide fumigation, development of treatment schedules for alternative fumigants such as sulfuryl fluoride has become a priority.

In China, crates, palettes, spools, and other wood packing items are often manufactured in small shops with limited budgets. Providing relatively simple, inexpensive, and effective methods of eliminating woodborers is an important component in ensuring compliance with 7CFR 319.4. A number of heat-based treatments, including conventional ovens and microwaves, are being developed to satisfy this need. APHIS-PPQ-CPHST, as the direct methods development arm of the regulatory agency, has taken the lead in working to meet this objective. CPHST and its U.S.-based cooperators are, in many cases, working directly with the Chinese on these projects.

Work Plans:
ARS: none
FS: none
APHIS: APHIS-02.12, APHIS-02.14, also see NOTE 2.

> *NOTE 2: APHIS-PPQ-CPHST has initiated an additional work plan in this area in response to the SWPM issue. The study goes beyond the scope of ALB and includes such pests as deep-wood pathogens. As a result, it is being funded from AQI user fees. CPHST and APHIS-IS are also conducting work to confirm the effectiveness of kiln heating (drying) for eliminating ALB in SWPM, in part through cooperative work with Chinese industries; this work was conducted using FY-2000 funds.*

OBJECTIVE AND JUSTIFICATON:
Improve methods for verification of regulatory treatments, including exclusion and timing of eradication actions.

Verification that wood used as SWPM was treated to kill ALB could, in some situations, be difficult. For example, while kiln drying substantially reduces the moisture content of wood, shorter-term heating (including microwave treatment) may leave little evidence of treatment, forcing APHIS inspectors to rely on written certification that wood was treated, perhaps in small business establishments that are physically removed from shipping ports. CPHST is working with cooperators to identify chemical indicators that can be applied to wood and change characteristics when heated to a range necessary to kill internal ALB.

Work Plans:
ARS: none
FS: none (related: FS 02.11)
APHIS: APHIS-02.14

OBJECTIVE AND JUSTIFICATION:
Strengthen public support and participation, especially from tree care professionals, for regulatory and exclusion activities.

This research focuses on a number of key needs: understanding of how homeowner beliefs affect implementation of current and future ALB control strategies; development of information and materials that facilitate communications by public personnel (regulatory officials and scientists) with homeowners; and, evaluate tree care professional preparedness for recognizing ALB infestation and susceptible trees.

Work plans:
ARS: none
Forest Service: FS 02.06; FS 02.07
APHIS: through appropriation to LPA, not to CPHST

C. CONTROL

Program

The ability to kill or otherwise block reproduction of an invading pest is critical for the success of eradication programs. The control methods of choice must be efficient, logistically feasible, and cost-effective. The social, health, and environmental consequences of pest control operations, along with public perception of those consequences, must also be considered when conducting eradication programs. Increasingly, the choice of control tactic to be used in a given situation is influenced by land-use patterns (e.g., agricultural vs. residential), environmental sensitivity of program areas, and other ecological, social, and political factors. In many cases, compromises exist between efficacy of a control method and its environmental/health safety and/or cost. As a result, programs have to take multi-faceted and locally tailored approaches to their control operations, and research and development groups strive to provide a variety of efficacious but environmentally benign control options.

For ALB, eradication programs still rely on removing and chipping trees as the control method of choice for eliminating known infestations. When trees are replanted at sites of tree removals, species that are not known ALB hosts are used, rendering the local environment unsuitable to the pest. Over the past two years, programs have also started treating uninfested trees with a systemic insecticide (imidicloprid) in an effort to protect those trees from ALB attack. This approach has less potential for impact on the environment than, say, broadcast sprays of broad-spectrum insecticides aimed at killing adult beetles (the latter approach has potential but has not been seriously considered due to problems inherent in aerial or other broadcast application of insecticides in the urban and suburban areas where the programs are being conducted). Further development of systemic insecticides is needed to ensure and enhance their efficacy for both preventative and, possibly, direct control of larvae in trees. With ALB, the relative inefficiency of current survey methods places a premium on effective controls and the availability of cost-effective and safe preventative (protective) control tactics. Such options as biologically based “insecticides” (pathogens) and semiochemical-based baits will be investigated in an effort to supply eradication programs with a safe but potent arsenal for eliminating this pest.

Goal: Develop novel control strategies and tools for eradication of ALB populations.

Approach: To facilitate rapid eradication of ALB, with minimal reinfestation of areas within and around the eradication zone, research will be conducted to develop insecticidal treatments (including chemicals and bio-insecticides such as fungi, bacteria, and protozoa) and to determine which species of ALB-resistant trees should be used to replace infested ALB-susceptible ones.

OBJECTIVE AND JUSTIFICATION:
Evaluate insecticides for efficacy against ALB larvae and adults.

Chemical control technology has been a mainstay of efforts to eradicate exotic agricultural and forest pests. For many insects, pesticides remain the surest, simplest, and most cost-effective control measure. For ALB, chemical control is not straightforward because the insect spends most of its life inside the wood of trees, where it is difficult to deliver insecticides. Based on early results by APHIS scientists and their Chinese cooperators, thousands of uninfested trees in New York and Illinois were given a protective treatment with the systemic insecticide imidicloprid this year; however, additional work is needed to optimize this tactic. In addition, use of spot sprays (or bait stations) is being investigated as a method of delivering insecticides to ALB adults while minimizing the amount and coverage of insecticide in the environment.

Testing of chemical compounds for control of ALB is being carried out by FS R&D, and methods development specialists in APHIS and FS S&PF. Work is being conducted in quarantine and in China. The principal aim of these studies is to test the effect of systemic insecticides on ALB mortality, and to evaluate the rate at which effective control occurs. Studies will also evaluate injection techniques. All chemical control testing is coordinated. The three groups involved, FS R&D, FS S&PF and APHIS are testing different materials on different host trees.

Availability of test organisms is a critical need for many components of the ALB R&D effort. As examples, work in the following areas has suffered from lack of available test insects: assessing toxicity of various insecticides to ALB larvae and adults, bioassays of candidate attractants, and understanding of various basic life-history parameters that influence pest risk assessments and management decisions.

Work plans:
ARS: none
Forest Service: FS 02.08; FS02.09; FS 02.11; FS 02.22, FS 02.25
APHIS: APHIS 02.01; APHIS 02.02; APHIS 02.03; APHIS 02.04; APHIS 02.05; APHIS 02.06; APHIS 02.07

OBJECTIVE AND JUSTIFICATION:
Evaluate biologically based controls.

Biological control includes such methods as the importation of parasitic wasps, predacious insects, and use of disease organisms (such as the fungus now suppressing gypsy moths).

ALB biologically based control options (e.g., microbial pesticides and biological control agents) are being evaluated. Moreover, these options are being explored as a means of providing additional tools for management within quarantine areas, wherein environmentally sensitive options are especially desired.

Scientists are conducting research on microbial control agents (fungi, bacteria, and protozoa) for ALB. ARS, FS, and APHIS, in collaboration with scientists at Cornell University, are evaluating promising fungi. FS scientists are identifying *Bacillus thuringiensis* (Bt) formulations, existing and custom-made, which are effective for control of ALB. Studies are also identifying pure Bt toxins that can be made into an effective biopesticide. And, FS scientists are identifying microorganisms associated with ALB and determining their relationship to ALB health. Numbers of protozoa (microsporidia) have also been isolated and positive tests have been obtained for Bt in the laboratory. These tests are being done both in quarantine and in China.

Because the focus is on eradication, and there is no cost-effective method for producing parasites and predators for mass release in biocontrol, this proposal does not specifically request support for evaluation of non-microbial biological control agents; however, preliminary evaluations are in progress with in-house support from ARS. This is being done with collaborators at the Chinese Academy of Forestry, the Northwest Forestry University in China, and the Inner Mongolia Academy of Forestry.

Work plans:
ARS: ARS 02.09
Forest Service**:** FS 02.01; FS02.02; FS02.03; FS 02.05; FS 02.08; FS 02.09; FS 02.15; FS 02.17; FS 02.18; FS 02.21, FS 02.26, FS 02.27
APHIS: APHIS 02.04; APHIS 02.07

OBJECTIVE AND JUSTIFICATION:
Evaluate tree species resistance.

The identification of resistant tree species is important for restoration of impacted areas and, also, for future control strategies should eradication fail. These investigations identify those tree species in the U.S. (urban and forest areas) that are preferentially attacked by ALB, and those tree species that are not attacked by ALB. This information will be used to determine what tree species should be used to replace those trees which have been cut and removed as a result of ALB attack, as well as what tree species should be used in reforesting urban and rural landscapes.

Scientists are also conducting research on host range of the ALB. In the U.S., most species of maple have been attacked, but also some species of poplar, willow, and elm, among others. In addition, in the U.S., three new tree genera have been reported to be attacked by ALB – horse chestnut, ash and Rose-of-Sharon. In mid-western and eastern US states, several poplar and willow species and hybrids are planted as short rotation woody crops, and several tree species are being used in New York and Chicago tree plantings. In addition to knowledge of host attractancy, study of host range can give vital information about the vulnerability of trees used for planting in urban areas and for short-rotation forestry.

Work plans:
ARS: ARS 02.10
Forest Service: FS 02.05
APHIS: APHIS 02.15; APHIS 02.16

OBJECTIVE AND JUSTIFICATION:
Develop Rearing Systems and Rear ALB in Support of Program Objectives.

Some basic methods for rearing ALB in a quarantine laboratory setting have been worked out, including multi-generation rearing and the development of several artificial diets for ALB grubs. However, ALB rearing remains extremely expensive and labor-intensive, and mortality of various life-stages is periodically much higher than desirable. Better, more efficient, methods are needed for all aspects of ALB rearing, including adult handling and feeding, oviposition and egg handling (including development of artificial oviposition substrates), larval maintenance, dormant (chill) periods, and pupation. In addition, rearing operations must continue to provide test organisms needed for the various R&D projects.

Work plans:
ARS: ARS 02.12
Forest Service: FS 02.12
APHIS: APHIS 02.17; APHIS 02.18

D. ERADICATION PROGRAM MANAGEMENT/OPERATIONS

Program

Large-scale eradication programs such as the ALB programs in New York and Illinois need effective means of tracking and prioritizing program operations. Survey and control efforts must be organized systematically in time and space in order to ensure that all areas are covered efficiently and without unnecessary duplication of effort. The potential effects of the insect on our tree resources must be understood in order to assess the cost-effectiveness of program options.

OBJECTIVE AND JUSTIFICATION:
Develop GIS/GPS data systems, and automated/remote data entry.

The ability to track locations of infestation foci and program activities such as control and survey operations in time and space is critical to the efficient operation of eradication programs. This research component includes development of a user-friendly Windows program that will allow municipalities to assess the potential impact and risk of the ALB to their urban forest resource. Using a simple sampling methodology, or an existing inventory of trees (e.g., street tree inventory), this program will determine: potential numbers of trees at risk to ALB infestation; total leaf area at risk to ALB infestation; total potential risk to urban forest sustainability; total risk to forest functions and benefits; total tree biomass and carbon at risk to ALB infestation; and, total monetary value of the resource at risk to ALB infestation.

Goal: Optimize eradication operations through improved forest data management systems and cost analyses.

Approach: Remote sensing and recording systems, together with automated remote data entry and information gleaned from other databases, will be used to develop maps and other analytical tools for prioritizing ALB eradication efforts. Simple methods to assess the urban forest resource, and, user-friendly computer programs to quantify the potential impact and risk to ALB infestation will be developed. An easy-to-use field manual and PDA (e.g., Palm Pilot) data-recording program will be developed to allow cities to easily sample or inventory their urban forest resource. This type of base data is necessary to assess the potential risk to the forest from ALB attack. When gathering these data, data collectors will also be monitoring for evidence of existing ALB populations. Data from these assessments will be entered into spreadsheets to serve as base data for the Windows ALB program. The user-friendly Windows desktop computer program will be developed to allow users to easily calculate the potential risk (and associated monetary value) of ALB infestation to their urban forest. Methods for field sampling and analysis will build on the existing Urban Forest Effects (UFORE) model methodology (Nowak and Crane, 2000) and ALB information (Nowak et al., 2001). This new computer program and users manual will allow local users a quick, easy, and low-cost method of analyzing their urban forest to help develop appropriate management strategies to protect their urban forest resource from ALB attack. Data analyzed in the program will be displayed in both tabular and graphic formats. The program will also allow for inventory data to be displayed within GIS mapping programs.

Work plans:
ARS: none
Forest Service: FS 02.23; FS 02.24
APHIS: APHIS 02.10; APHIS 02.11

OBJECTIVE AND JUSTIFICATION:
Evaluate current and/or potential impacts.

Research will build upon an assessment of the potential impact of ALB in urban areas of the U.S. (Nowak et al., 2001). This work will analyze the potential impact of ALB on U.S. forests nationwide. For each U.S. county, the total (and percent) number of trees and basal area that is susceptible to ALB attack will be calculated. Forest data to determine susceptibility to ALB will be derived from the FIA national database. From these data, information will be derived to determine the compensatory value of the forest, using four variables (species, diameter, condition and location of the forest). Local species and diameter specific values used in the formula are derived from state value. Compensatory value loss due to potential ALB impact will be calculated for each county.

Range limitations will be determined using average maximum and minimum temperatures, and annual precipitation data will be compiled for each county. These data will be used to help display potential range limitations to ALB infestation in the United States.

GIS maps will be developed that display national ALB susceptibility data will be developed using Arc View. These GIS maps will be used to display, by county, U.S. forest information on: proportion and total number of trees susceptible to ALB; proportion and total basal area susceptible to ALB; total compensatory value of trees susceptible to ALB; and average maximum and minimum temperatures, and annual precipitation (these data can be used to develop potential range limitation maps for ALB).

Work plans:
ARS: none
Forest Service: FS 02.23; FS 02.24
APHIS: APHIS 02.13; APHIS 02.15; APHIS 02.16

Appendix

Agency Work plans

USDA ARS Project Descriptions

Work plan Number: ARS 02.01

Project Title: ALB Attractants and Traps

Principal Investigators & Affiliation: J. Aldrich, A. Zhang, USDA ARS Chemical Affecting Insect Behavior Laboratory, Beltsville, MD

Other Cooperators: Dr. Zhao, Dr. Xia, Trecé Corporation,

Project Objectives and Approach: (1) Identify new compounds and (2) develop traps for ALB.

Project Deliverables: Effective means of detecting ALB populations, with possible additional application to beetle population reduction.

Update: ARS scientists have isolated, identified, and synthesized two male-specific compounds from the ALB that attract walking females and males (patent US 6,177,073 B1); these are the first attractants identified from ALB, proving that this line of investigation is feasible. Additional chemical cues are needed, e.g., ones effective over longer distances.

Work plan Number: ARS 02.02

Project Title: *Anoplophora* Systematics and Expert Identification Systems

Principal Investigators & Affiliation: S. Lingafelter, USDA-ARS Systematics Entomology Laboratory, Beltsville, MD

Other Cooperators: APHIS port operations would provide insects

Project Objectives and Approach: (1) Conduct *Anoplophora* systematics and (2) develop expert identification systems.

Project Deliverables: Relationship (cladogram) analyses and expert identification systems for *Anoplophora* species that uses morphological and molecular characters. This will not only improve port identifications, but will also provide additional relationship-based clues to biology, including susceptibility to attractants, that will be useful in detection and in developing eradication strategies. This will improve chances of intercepting and treating related pest beetle species, and will decrease expenditures of valuable quarantine and eradication funds, and concomitant removal of valued street, yard, and park trees and environmental damage due to insecticide exposure, during treatment of non-pest beetles. The development of the expert identification systems will be applicable to interdiction of other potential invasive species.

Update: ARS scientists have produced a systematics monograph (2001 publication date) of ALB and 40 species of *Anoplophora*; many are serious potential pests that are easily transported in wood. The monograph has descriptions, keys, photographs, and descriptions of variation.

Work plan Number: ARS 02.03

Project Title: ALB Dispersal and Movement Behavior

Principal Investigators & Affiliation: M. Smith and J. Bancroft, USDA-ARS, Beneficial Insects Introduction Research Laboratory, Newark, DE

Other Cooperators: Gao Ruitong, Chinese Academy of Forestry, Beijing, PRC; APHIS ALB eradication personnel in New York City and Chicago

Project Objectives and Approach: (1) Determine ALB behavior, specifically population dispersal and individual movement. Key variables that influence beetle dispersal and movement, together with ALB reproductive capacity (see Update below), will be used to develop a dispersal model, which will be used as a population model to predict beetle abundance and spread. This model will be tested by comparing predictions with ALB abundance from the field, and will then be applied to U.S. infestations to create a probability of presence (risk map) for ALB. This requires a prediction of beetle distribution in sites of U.S. infestations. We plan to coordinate with APHIS officials in order to acquire data on quarantine surveillance effort, tree removal, and pesticide treatment. Surveillance effort data would be used to evaluate the relative cost and efficiency of the three surveillance methods currently utilized (ground, bucket truck, tree climber). Tree removal data will be used to provide estimates of beetle populations and in the simulations. The cost and effectiveness of the systemic insecticide imidacloprid is needed to optimize adaptive eradication strategies.

Project Deliverables: The resulting models will provide APHIS with landscape-specific recommendations for exactly where and how resources should be used to efficiently eradicate ALB. This will greatly increase cost-effectiveness of detection and survey efforts used in the eradication program. Furthermore, the resulting models will allow APHIS to identify host-trees within the ambit of dispersal from individually infested trees, to select appropriate survey methods, and to focus survey efforts. Finally, the information developed in these studies will aid APHIS in the operational aspects of treatment application to individual trees.

Project Update: ARS scientists, in cooperation with scientists at the Chinese Academy of Forestry, are studying ALB in infested landscapes in China analogous to urban landscapes in the U.S. Using population level mass mark recapture methodology, in which ca. 20,000 ALB were marked and released during both 1999 and 2000, these studies have shown that ALB is capable of flying over 2,600 m in a single season, which resulted in modifications in the APHIS survey and containment perimeters. ARS now has: (1) two years of data on ALB seasonal abundance and dispersal capacity; (2) over 1,000 observations of the daily movement (distance and propensity) of individual beetles in trees similar in structure to urban trees; (3) over 1,000 observations on the instantaneous flight capacity of individual beetles; and (4) detailed data on the propensity of individual beetles to remain in or depart from their natal host tree (propensity). Key variables that influence beetle dispersal and movement (*i.e.,* tree characteristics, tree health, landscape characteristics, beetle sex, size and age, and environmental variables) have been quantified in these studies to facilitate the prediction of movement in urban landscapes currently infested by ALB (first paper has been accepted by referred scientific journal for publication; scheduled publishing in 2001). ARS has also completed studies of the reproductive capacity of ALB on several of the most commonly infested host trees in the U.S., including Norway maple (first paper has been accepted by referred scientific journal for publication; scheduled publishing in 2001).

Work plan Number: ARS 02.04

Project Title: Acoustic Signature Identification of Feeding ALB Larvae

Principal Investigators & Affiliation: M. Smith, USDA-ARS, Beneficial Insects Introduction Research Laboratory, Newark, DE

Other Cooperators: S. Teale, State University of New York at Syracuse; Pryor Knowledge Systems (PKS), Inc.

Project Objectives and Approach: (1) Develop and verify a neural network-based acoustic signature system for detecting ALB larvae feeding in trees in urban areas, as well as in cargo (pallets, dunnage, bonsai, etc.).

Project Deliverables: Together with a detector, this will yield a ground-based system that can detect ALB feeding in the canopies of trees, and that is also applicable to detecting the beetle in cargo.

Update: Since 1998, ARS scientists (Michael T. Smith, Research Entomologist, BIIRL) in collaboration with scientists at the State University of New York at Syracuse, and acoustical technology researchers at Pryor Knowledge Systems, Inc. (PKSI) (a company that has successfully developed innovative acoustic learning systems, particularly in noisy environments), have been developing acoustical technologies for the detection of ALB larvae feeding within trees, showing that it is possible to detect larvae feeding within live poplar, willow, and elm trees under natural field conditions where there is significant background noise. Furthermore, these feeding sounds have been detected up to 18 feet above the ground, and within the three primary tissues within trees where ALB larvae feed. More recently, working in the New York ALB Quarantine Area, the acoustical technology has also been shown to detect ALB larvae (1st to last instar) feeding in the same tissues in Norway maple (one of the most common species attacked by ALB in the U.S.), and specifically within this very noisy urban landscape. Therefore, this research holds high potential for allowing early and accurate detection of ALB-infested trees.

Work plan Number: ARS 02.05

Project Title: Lightweight Acoustic Detection Kit for ALB Larvae w/ Signal Interpretation by Human Listeners

Principal Investigators & Affiliation: M. Smith, USDA-ARS, Beneficial Insects Introduction Research Laboratory, Newark, DE

Other Cooperators: S. Teale, State University of New York at Syracuse; Pryor Knowledge Systems (PKS), Inc.

Project Objectives and Approach: Rapid development of a lightweight ALB detection kit, using human "trained ear" interpretation of sounds. This kit will comprise a lightweight electronic ALB Detector, with earphones, and a lightweight electronic player that replays previously recorded ALB feeding sounds to train the ear of the person using the kit. This would be a 'passive' system, which listens for ALB larval feeding sounds. Two contact transducers will be considered: one invasive and one non-invasive. (1) In Phase 1, the acoustical signatures of ALB larval feeding will be defined, and (2) the prototype will be refined to allow several replicas to be built by PKS, Inc. in Phase 2, with field-testing by ARS. The goal of Phase 2 would be to provide the system replicas to APHIS for field-testing of the system within the framework of the eradication surveillance program. (3) Feedback from field-testing results by APHIS and their subcontractors will be used to determine whether subsequent modifications to the ALB Detection Kit will be needed before producing the kit in large volumes (Phase 3) for final field distribution.

Project Deliverables: This kit will be a readily available interim solution suitable for easy deployment to field agents for the critical inspection of infested trees, imported wood products, bonsai trees, crating, and pallets. It is not a complete substitute for a computer-based detection system in large live trees.

Update: See Work plan Number ARS 02-04.

Work plan Number: ARS 02.06

Project Title: Treetop Acoustic Detection Kit for ALB w/ Signals Interpreted by Computer Software

Principal Investigators & Affiliation: M. Smith, USDA-ARS, Beneficial Insects Introduction Research Laboratory, Newark, DE

Other Cooperators: Pryor Knowledge Systems (PKS), Inc.

Project Objectives and Approach: (1) Develop an electronic long distance ALB detector, with earphones, and an electronic signal output suitable for input to a recording device or computer. See Work plan Number ARS 02-05 for Approach.

Project Deliverables: This system will allow survey personnel to monitor adult ALB feeding on leaves and twigs and adult female ALB gnawing of oviposition sites in trees during the window of time that ALB adults are active, which tends to be primarily from mid June to early September. The desired goal for time to completion on this project would be for the July 2002 research season in the U.S. and China. However, such a system could be delivered in approximately 4 months from the time of funding.

Update: See Work plan Number ARS 02-06

Work plan Number: ARS 02.07

Project Title: Ground Level Acoustic Detection Kit for ALB w/ Signals Interpreted by Computer Software

Principal Investigators & Affiliation: M. Smith, USDA-ARS, Beneficial Insects Introduction Research Laboratory, Newark, DE

Other Cooperators: Pryor Knowledge Systems (PKS), Inc.

Project Objectives and Approach: (1) Design a non-invasive ground-level electronic ALB feeding detection and monitoring system to detect ALB larval feeding sounds in tree-top branches and the trunk from ground-level. This would be andactive' system in which trees would be illuminated and information then collected via a receiver. See Work plan Number ARS 02-05 for Approach.

Project Deliverables: A ground-based detection system with computer signal interpretation that would be man-portable and not damage the bark of the tree. The system should be capable of automatically distinguishing ALB feeding sounds from other ambient noises.

Update: See Work plan Number ARS 02-04.

Work plan Number: ARS 02.08

Project Title: ALB Visual Cues for Developing ALB Monitoring Systems

Principal Investigators & Affiliation: M. Smith, USDA-ARS, Beneficial Insects Introduction Research Laboratory, Newark, DE

Other Cooperators: Gao Ruitong, Chinese Academy of Forestry, Beijing, PRC

Project Objectives and Approach: (1) Determine the visual cues used by ALB in host location, orientation and selection, and identify visual cues that can be used in beetle monitoring. These studies include evaluation of beetle (sex and age), host tree (physical characteristics and health), landscape characteristics (qualitative and spatial), and environmental factors (*e.g.,* temperature, humidity, wind) on ALB host selection. Studies will also assess short and long-range host orientation behavior. Studies will be conducted in urban and forest settings to determine if urban background (visual) noise influences the use of visual and/or olfactory signals.

Project Deliverables: Identification of ALB visual cues that can be used to improve trap design for ALB monitoring, as well as in development of sentinel and/or bait tree methods as survey and control strategies.

Project Update: Chemical odor cues have been shown by ARS scientists to attract walking male and female beetles. However, ARS scientists, working in collaboration with colleagues at the Chinese Academy of Forestry, have been conducting field studies and showed that visual cues also play a major role in host tree orientation and selection behavior. ARS now has several years of detailed data on in-flight host orientation behavior, specifically in reference to key environmental and landscape factors. Studies will continue, with an emphasis on beetle, host tree and landscape spatial factors, and their influence on ALB host selection. The goals of these studies are to aid in development of trapping systems (design and guidelines), and the development of sentinel trees and/or bait trees for monitoring and control methods.

Related Approach: Survey: 1) Host Location Mechanisms; 3) Developing a Trapping System.

Work plan Number: ARS 02.09

Project Title: Fungal Control of ALB

Principal Investigators & Affiliation: J. Vandenberg, USDA-ARS, Plant Protection Research Laboratory, Ithaca, NY; H. Chen, USDA-ARS Sino-American Biological Control Laboratory, Beijing, China; Frank Herard and William Meikle, USDA-ARS European Biological Control Laboratory, Montpellier, France; M. Smith, USDA-ARS, Beneficial Insects Introduction Research Laboratory, Newark, DE

Other Cooperators: Ann Hajek, Cornell University.

Project Objectives and Approach: (1) Develop fungal pathogens for ALB eradication. The collaboration with Cornell will be continued, with focus on fungal pathogens, and sub-objectives to: (a) develop methods for growing fungal strains on bands and determine whether strains differ significantly in growth characteristics, *e.g.,* density of spores produced, focusing on the maximally virulent fungal strain(s); (b) determine how long bands are effective in the field by conducting bioassays in quarantine facilities using bands from the field, with comparisons of persistence among the most virulent fungal strains; and (c) optimize band placement to maximize the persistence and activity of the fungus in bands in the field, while also optimizing band position to improve chances of contacting searching beetles. In addition to collections and testing of fungi from China by Dr. Hajek, the ARS European Biological Control Lab will collect and evaluate pathogens of cerambycids native to Europe as candidate pathogens for use against ALB (complementing the FS focus on bacteria, nematodes, and protozoa). In the U.S., pathogen effectiveness will be evaluated at the ARS-Ithaca quarantine facility, as is currently being done by Dr. Hajek.

Project Deliverables: Eradication of ALB using fungi in an inundative microbial control approach. Fungal pathogens are demonstrating excellent promise for biological control of ALB. A non-woven fiber band impregnated with the insect pathogenic fungus *Beauveria brongniartii* is presently for sale in Japan for controlling a closely related longhorned beetle. Once attached to tree trunks and branches, wandering adult beetles contact the bands and become inoculated with the fungal spores. This unique application technique may be especially well suited to controlling ALB because the fungus in bands remains active in Japan for 30 days, as it dries and re-wets each night to produce more spores. By comparison, spores of insect pathogenic fungi applied as a foliar spray do not survive more than a few days. Continued studies investigating use of this technology toward eradication of ALB are needed. Furthermore, an insect pathogenic fungus will be particularly appropriate for ALB control since biological control is preferred for use in urban areas where ALB occurs.

Update: Biological control of ALB may be a useful approach for long-term management of ALB; however, for eradication, the inundative use of microbes would be a more effective approach. That is, microbes, once identified, can be sprayed on trees and/or applied to a specialized substrate that is attached to trees in order to complement chemical treatments. In particular, microbes could be used adjacent to the current ALB containment area to provide additional protection provided by systemic insecticides such as imidacloprid (which have been shown to kill 50% of adult and larval beetles contacting it). This collaborative project with researchers at Cornell University was funded by an emergency allocation that expired in 2000. Field trials comparing fungal sprays versus bands using two different fungal strains showed that bands with *B. brongniartii*, taking advantage of field observations of the searching behavior of the beetle (See details under Host Colonization Behavior), were superior both in reducing oviposition and in speeding mortality of adults. Bioassays with different fungal strains suggest that new fungal strains (including some from the U.S.) could be more effective than the commercially available strain. (See details under Host Colonization Behavior). Using this technique, Japanese colleagues have successfully controlled *Anoplophora malasiaca* in citrus.

Work plan Number: ARS 02.10

Project Title: Resistant Trees for Urban Street Plantings

Principal Investigators & Affiliation: M. Smith, USDA-ARS, Beneficial Insects Introduction Research Laboratory, Newark, DE; H. Chen, USDA-ARS Sino-American Biological Control Laboratory, Beijing, China

Other Cooperators: K. Hoover, Pennsylvania State University, State College, PA

Project Objectives and Approach: (1) Identify trees resistant to ALB for use in urban areas. (2) Evaluate the role of tree stress in host susceptibility.
Project Deliverables: Resistant trees suitable for planting in eradication zones will be identified.

Update: To facilitate eradication, removal of infested trees should be accompanied by the planting of trees resistant to infestation. In the U.S., most species of maple have been attacked, as have some species of ash, birch, poplar, willow, sycamore, basswood, cherry, pear, elm and horse chestnut (this includes three previously unreported tree genera attacked by ALB -- horse chestnut, ash, and Rose-of-Sharon). ARS has developed methodology and completed studies of the reproductive capacity of ALB on several of the infested host trees in the U.S., including Norway maple (first paper has been accepted by referred scientific journal for publication; scheduled publishing in 2001). ARS has also evaluated host suitability, measured by larval weight gain, among eight tree species in the U.S. (Chinese elm, Norway maple, American elm, honey locust, sugar maple, red oak, white ash, green ash), specifically as a function of natal host. These studies suggest that concentrating survey efforts on species of maple is appropriate for eradication, and that honey locust should not be used for replanting impacted areas (first paper has been submitted for publication in a refereed scientific journal).

Work plan Number: ARS 02.11

Project Title: ALB Host Colonization Behaviors (Mating, Oviposition and Feeding)

Principal Investigators & Affiliation: M. Smith and J. Bancroft, USDA-ARS, Beneficial Insects Introduction Research Laboratory, Newark, DE

Other Cooperators: G. Ruitong, Chinese Academy of Forestry, Beijing, PRC.

Project Objectives and Approach: (1) Determine ALB colonization behaviors, including mating (*e.g.,* mate location), ovipositional and feeding behaviors (includes temporal and spatial qualities); and (2) Identify and quantify variables which influence beetle mating, ovipositional and feeding behavior, so as to enable prediction of these behaviors in a wide variety of infested areas, including those currently infested in the U.S.

Project Deliverables: The resulting information, together with ALB reproductive capacity, will aid in the development of monitoring (survey) methods and identify key behaviors that can be exploited in development of control strategies. Information from these studies, specifically within-tree searching behavior, has already been utilized by collaborators working on the delivery of entomopathogens for control of ALB within individual trees. The resulting information will also enhance improvement of visual survey methods, particularly early detection. Information from these studies, specifically ovipositional behavior, is already being utilized to develop more descriptive guidelines for the visual appearance of oviposition scars, which are a key sign of attack that is crucial for early detection. Furthermore, information from these studies will provide the behavioral template for investigations of host range and host preference, and will be incorporated into the dispersal model being developed (See details under Dispersal and Behavior).

Update: ARS scientists, in cooperation with scientists at the Chinese Academy of Forestry, are studying ALB behavior on infested landscapes in China analogous to urban landscapes in the U.S. ARS now has several years of detailed data on individual beetle behavior within trees, including mating and oviposition. Key variables that influence beetle behavior, particularly mating and oviposition (*i.e.,* tree characteristics, tree health, landscape characteristics, beetle sex, size and age, and environmental variables) are being quantified in these studies so as to identify key aspects of ALB behavior that can be exploited in the development of monitoring and control methods. ARS also now has over 5,000 observations on the visual appearance of ovipositional scars among various tree species, including Norway maple, and has shown that the visual appearance differs among tree species (first paper is in preparation and will be submitted to a referred scientific journal for publication during 2001).

Related Projects: A. Survey, 1) Develop a Trapping System; Improve Current Visual Survey Methods and Technology; C. Control, 2) Biological Control; E. Rearing and Fundamental Biology, 2) Host and Geographic Range, 3) ALB Colonization Behaviors.

Work plan Number: ARS 02.12

Project Title: Rearing of ALB

Principal Investigators & Affiliation: M. Smith, USDA-ARS, Beneficial Insects Introduction Research Laboratory, Newark, DE

Other Cooperators: Ann Hajek, Cornell University.

Project Objectives and Approach: (1) Develop methods for rearing ALB. The collaboration with Cornell will be continued, with focus on: (a) evaluation of larval diapause and determining at what time in development larvae will respond to a chill, stop developing, and enter diapause; (b) determining the length of the required chill period and what conditions optimize survival during the chill period, *e.g.,* instar and moisture level; and (c) investigating whether specific photoperiods shorten diapause length or synchronize diapause termination.

Project Deliverables: Researchers must often wait for prolonged periods of time in order to work with later instar ALB and adults due to the slow development of the beetles. Also, lack of understanding of ALB larval diapause means that researchers could be waiting longer than necessary for larval diapause to terminate. The proposed studies will continue to optimize the rearing protocol for ALB, so that only those larvae needing to diapause will be put into chill, all larvae will survive during chill, and the larvae will be in chill for an optimal length of time (maximizing survival as well as subsequent eclosion to adult).

Update: A. Hajek, Cornell University, has studied Japanese species of cerambycids and has demonstrated that photoperiod can influence diapause.

USDA Forest Service R&D Project Descriptions

<u>Work plan Number:</u> FS 02.01   <u>Goal</u>: Control

<u>Project Title:</u> Use of Systemic Antibiotics for Control of *Anoplophora glabripennis* by Targeting Midgut Symbionts

<u>Principal Investigator(s) name, location and affiliation:</u>
Leah Bauer, Forest Service, North Central Research Station, E. Lansing, MI
Deborah Miller, Forest Service, North Central Research Station, E. Lansing, MI
Ken Raffa, University of Wisconsin, Madison, WI

<u>Project Objectives/Approach:</u>
Identify bacterial symbionts responsible for digestion of cellulose in the midgut of *A. glabripennis* and evaluate the use of antibiotics for control of larval stages.

<u>Research Approach:</u>
During 2002, scientists will identify and compare midgut symbionts in *A. glabripennis* larvae reared on diet and in larvae collected from different host tree species. Antibiotics will be evaluated for control of larvae by the diet incorporation method, followed by comparisons of larval control by injection into host trees. The impact of antibiotics on midgut symbionts will be evaluated post-treatment.

<u>Project Deliverables:</u>
The use of antibiotics, some of which are small peptides, may provide alternatives to conventional pesticides for control of *A. glabripennis* by targeting the midgut symbionts responsible for digesting the cellulose. These compounds may have less impact on non-target species such as predatory insects and birds.

<u>Work Plan Number</u>: FS 02.02   <u>Goal</u>: Control

<u>Project Title:</u> Survey and evaluate Entomopathogens of *Anoplophora glabripennis* (Motschulsky).

<u>Principal Investigator(s) name, location and affiliation:</u>
Leah Bauer, Forest Service, North Central Research Station, E. Lansing, MI
lbauer/nc_el@fs.fed.us
Deborah Miller, Forest Service, North Central Research Station, E. Lansing, MI
dmiller/nc_el@fs.fed.us
Houping Lui, Michigan State University, E. Lansing, MI
Ann Hajek, Cornell University, Ithaca, NY
Rich Humber, USDA-ARS, Ithaca, NY
Charles Vossbrinck, Conn. Ag. Exp. Station, New Haven, CN
Ke Dong, Depts. Entomol. & CIPS, MSU, E. Lansing, MI; dongk@msu.edu
Tonghia Zhao, Beijing Academy of Forestry, Beijing, China; zhaoth@mail.forestry.ac.cn

<u>Technical Staff: name, location, and affiliation:</u>

Amy Francisco, USDA FS NCRS, E. Lansing, MI; francisco@msu.edu
Ann Christiensen, Dept. Entomol., MSU, E. Lansing, MI; chris263@msu.edu

Project Objectives/Approach:
(Objective 1) Survey populations of larval and adult *A. glabripennis* for entomopathogens in Asia. During 2002, scientists will travel to Asian countries where *A. glabripennis* is endemic. Life stages will be collected for propagation of live microorganisms, transmission electron microscopy, and for genetic analyses. Potential entomopathogens will be evaluated for infectivity and impact on A. glabripennis in North America.

(Objective 2) Evaluate entomopathogens for utility for biocontrol. This study is comprised of several components: (a) Isolate and identify entomopathogens from ALB in quarantine laboratories in North America and from collections made in China for bioassays in ALB and other cerambycids; (b) Bioassay protein toxins, including those from *Bacillus thuringiensis*, in ALB and other cerambycids; (c) Prepare ALB midguts for development of an in vitro toxin bioassay in collaboration with other researchers.

Project Update: Scientists collected ALB from field populations and continue to receive cadavers from ALB quarantine laboratories. While working in China in October 2000, they collected dead and live ALB larvae and adults in water. Cooperators at the Chinese Academy of Forestry (CAF) blended the ALB and washed the homogenate by centrifugation. In January 2001, they received the ALB homogenate from China, but must purify and isolate fractions before inoculating our surrogate species, the cottonwood borer (CWB), with suspect particles. Entomopathogens discovered to date include several isolates of microsporidia and fungi; microsporidia are being studied in CWB after per os inoculation, studied using transmission electron microscopy, Giemsa smears, and taxonomic and genetic status will be done in collaboration with Lee Solter (INHS) and Charles Vossbrinck (CT Ag Exp Sta), respectively; fungal studies and identifications are being done in collaboration with Ann Hajek (Cornell Univ.) and Rich Humber (ARS, Ithaca); identification of bacteria is being done in collaboration with John Podgewaite (NERS, Hamden); Bt in vivo bioassays are done at the FS lab in E. Lansing using CWB; and, principle investigators are collaborating with Al Valaitis (NERS, Delaware) on the Bt project to develop an in vitro Bt assay using midgut tissue from CWB and ALB. No viruses are confirmed from quarantine or field samples, although some are described in the literature.

(Objective 3): Evaluate Asian longhorned beetle, Anoplophora glabripennis (Motschulsky), entomopathogens using a native cerambycid, the cottonwood borer, Plectrodera scalator (Fabricius), as a surrogate host. This objective requires three efforts: (a) Establish a colony of cottonwood borers (CWB); (b) inoculate CWB with entomopathogens isolated from ALB and determine infectivity and impact on the host; (c) use CWB to produce pathogens for bioassay of Asian longhorned beetle (ALB) at quarantine laboratories.

Interim findings. About 70 CWB adults were collected from cottonwood and willow in Michigan during late summer 2000. Thirty mating pairs oviposited in willow logs over a 3-month period. After various experimental trials, we determined that optimal laboratory rearing conditions include: exposure of the egg in the oviposition pit using a knife; unglue egg with a mild solution of NaOH; surface sterilize eggs for 10 min in 10% neutral buffered formalin; after eclosion, place neonates individually in pit cut into surface of artificial diet (modified Prionis) in 2 oz plastic cup; larvae are placed on fresh diet every 3 weeks into 4 and 8 oz wide mouth jars. Using this method, scientists reared over 500 larvae with minimal contamination and mortality. As this population was larger than necessary to support a laboratory colony, we chilled half for later use, while others are being used by other scientists. For example, inoculation of CWB with ALB microsporidia; bioassay with Bt toxins; diet-incorporation bioassays to estimate the CWB LC_{50} of insecticides for use in ALB studies at quarantine labs; transplantation of CWB into logs for use acoustic studies in E. Lansing; CWB were shipped to PSU at University Park for microwave studies.

The CWB appears to be a useful surrogate model for ALB due to its high genetic relatedness, similar behavior, host range, size, and response to artificial diet, and other laboratory conditions. Because it is native, quarantine facilities are not required. Moreover, CWB may be useful in the study of non-target insects should biological controls be implemented. Although this research is in progress, some of our work with CWB is already being applied to ALB studies and rearing protocols.

<u>Project Deliverable:</u> The introduction and establishment of natural enemies in N. America is an essential tool for mitigation of *A. glabripennis* impacts.

<u>Work plan Number:</u> FS02.03 <u>Goal</u>: Control

<u>Project Title:</u> Determine the Infectivity of *Beauveria* spp. to non-target insects in North America

<u>Principal Investigator(s) name, location and affiliation:</u>
Leah Bauer, Forest Service, North Central Research Station, E. Lansing, MI
Deborah Miller, Forest Service, North Central Research Station, E. Lansing, MI
Ann Hajek, Cornell University, Ithaca, NY

<u>Project Objectives/Approach:</u>
To determine infectivity of *Beauveria* sp., used for control of related citrus cerambycids in Japan, in non-target species in N. America.

During 2002, scientists will collect and rear North American cerambycids. Larvae and adults will be screen with *Beauveria* bands for infectivity to this fungus, as used operationally on a related citrus cerambycid in Japan. They will test at least 12 species of native cerambycids. They will also determine the genetic relatedness of the *Beauveria* sp. used in bands, with other *Beauveria* spp. native to North America.

<u>Project Deliverables:</u>

The use of commercially available *Beauveria* bands in North American *A. glabripennis* infestations is expected to slow the rate of spread of ALB, especially near present infestations.

Work plan Number: FS02.04 Goal: Survey; and, Regulatory/Exclusion

Project Title: Volatile Detection of Asian Longhorned Beetle Larvae (New)

Principal Investigator(s) name, location and affiliation:
Robert Haack, Forest Service, North Central Research Station, E. Lansing, MI; rhaack@fs.fed.us
Therese Poland, Forest Service, North Central Research Station, E. Lansing, MI; tpoland@fs.fed.us
Toby Petrice, Forest Service, North Central Research Station, E. Lansing, MI; tpetrice@fs.fed.us

Other Cooperators:
Oak Ridge National Laboratory, Organic Chemistry Division, Oak Ridge, TN
Center for Natural Products Chemistry, Michigan State University, E. Lansing, MI

Project Objective:
To identify unique volatile organic chemicals (VOC's) associated with the presence of the Asian longhorned beetle in live trees and in solid wood packing materials.

Research Approach: Working with staff at the Oak Ridge National Laboratory and Michigan state University, scientists will collect, analyze, and identify chemicals associated with ALB larval feeding sites on trees and in wood, including frass and sap exudate from the feeding sites. There is a high likelihood that some of these compounds will be unique to ALB and therefore represent a chemical signature of the organism. Scientists will collect compounds using various techniques, including solid phase molecular extraction, in both the US and in China. Compounds will be identified using coupled gas chromatography/mass spectrometry. Scientists will also collect compounds from uninfested trees/wood as well as trees/wood that are infested other species of woodborers.

Benefits: Currently, all infested trees are discovered through visual inspection, including the use of tree climbers and bucket trucks. Similarly, at US ports-of-entry, visual inspection is the only method used to detect infested solid wood packing material. Using advanced chemical-detection technologies would greatly improve the chances for early detection of infested trees and wood packing. Such technology would benefit USDA APHIS in their current ALB detection and eradication program, as well as in their monitoring programs at all US ports-of-entry.

Work plan Number: FS02.05 Goal: Program Management; and, Control

Project Title: Host Range of Asian Longhorned Beetle, and Evaluation of Poplar Resistance

Principal Investigator(s) name, location and affiliation:
Robert Haack, Forest Service, North Central Research Station, E. Lansing, MI; rhaack@fs.fed.us
Therese Poland, Forest Service, North Central Research Station, E. Lansing, MI; tpoland@fs.fed.us
Dr. Melody Keena, USDA Forest Service, Northeastern Experiment Station, Ansonia, CT; mkeena@fs.fed.us
Toby Petrice, Forest Service, North Central Research Station, E. Lansing, MI; tpetrice@fs.fed.us

Other Cooperators:
Deborah McCullough, Michigan State University, Entomology, E. Lansing, MI, mccullod@msue.msu.edu
Laura Lazarus, Michigan State University, Entomology, E. Lansing, MI lazarusl@pilot.msu.edu
Chinese Academy of Forestry and Beijing Forestry University

Project Objective/Approach:

(Objective 1) host selection behavior and suitability of several hardwood tree species now being used for replanting in Chicago and New York for ALB development. ALB is known to preferentially attack maples, elm, and willow in China, but new hosts such as ash, horse chestnut, and Rose-of-Sharon have been recorded in North America. Our objectives were to evaluate the suitability of selected North American hardwood species for oviposition by adult female beetles and development of larvae. Species of interest include economically important trees in the north central and northeastern U.S. and species used for replanting following eradication activities in New York and Chicago. A second objective was to assess behaviors associated with host selection by adult females.

Update: In 20002001, we worked at the Forest Service lab in Ansonia to test suitability of eastern cottonwood, honey locust, northern red oak, white oak, sycamore and tulip tree for larval development. Sugar maple, known to be highly preferred, was used for comparison purposes. We were provided with 8 females; each female was allowed to oviposit directly on a cut branch sample of one of the species for four days. Females were provided with sugar maple twigs for regeneration feeding. Branch samples were dissected after 21 days and developing larvae were counted. A few first instar larvae survived on both oak species and honey locust, but there were no larvae on eastern cottonwood, sycamore, or tulip tree.

We developed a four-armed olfactory chamber to evaluate host preference and host selection behaviors of adult, virgin ALB females. We tested eastern cottonwood, honey locust, northern red oak, white oak, sycamore and tulip tree, and compared them with sugar maple. A section of wood with bark attached, plus 7-10 twigs, was suspended in two of the four arms. Sugar maple sections and twigs were placed in the other two arms. Duration of all behaviors during a 50-minute interval was determined by observation and videotaping. Preliminary results show that sugar maple was significantly more attractive than alternate test species. Northern red oak appeared to be somewhat attractive, but other species were not.

Host selection behavior was further assessed by monitoring a female beetle for a 24 hr period in the olfactory chamber to assess types, duration, and sequences of behaviors. Sugar maple sections and twigs were suspended from two of the chamber arms and the other two arms were left empty. This was repeated with 3 different beetles.

In FY 2002, scientists will repeat host selection, host suitability and behavioral studies with additional ALB adults that have emerged from infested wood. If healthy ALB adults will again be scarce, scientists will work with native beetles as “surrogates” for ALB. Scientists have begun to share data and plan to pursue further collaboration with other scientists interested in host selection questions.

(Objective 2): Evaluate the susceptibility of the most commonly planted hybrid poplars and willows in the Great Lakes region to ALB attack; and, the mechanisms of tree resistance to ALB attack.

Several poplar species have been reported to be highly resistant to ALB in China, including *P. alba* var. *tomentosa* and *P. pekinensis*. Earlier studies by Chinese scientists have suggested that the mechanisms of resistance may be related to foliar pubescence, presence of stone cells within the bark, enhanced wound response, and high bark water content. More detailed studies will be conducted in China, using several species of Chinese poplars that vary in their resistance to ALB. Initially, we will evaluate ALB adult feeding, oviposition, and larval survival on all test trees. Scientists will measure and compare several foliar, twig, and wood characteristics in relation to ALB success on each poplar species. Cuttings from the most highly susceptible and resistant poplars will identify and propagated for further study into the genetic basis for resistance.

Update: In 1999, we tested several tree species, including species of Acer, Celtis, Corylus, Ostrya, Populus, Tilia, and Ulmus. In 2000/2001, cooperators at Michigan State University took the lead on testing the suitability of several hardwood tree species that are now used for replanting in Chicago and New York.

Scientists initiated work to plant cuttings from fast-growing hybrids poplars in the US in China and then test their susceptibility to ALB attack. Guidelines for movement of plant material to China were sought, and a search in China for already-established US hybrid poplar clones was requested. This work is ongoing. The best hybrid clones now being planted in the Midwest were identified as well as plantations where cuttings could be collected. Two planting sites have been identified in China, one near Beijing and one in Gansu Province.

Scientists observed oviposition on some tree species now being used for tree planting, e.g., Tilia. In 2000/2001 studies that were conducted by our cooperator, Quercus was used for oviposition in the lab. In both of these studies, few host species were presented to the adults, and so these results may not reflect what will happen in nature.

In FY 2002, scientists propose comparing lab results on cut branch samples to studies by others where live trees are being used, e.g., the work now being conducted at Penn State University or in China. Work is also proposed in 2002 at the Canadian Forest Service quarantine laboratory in Sault Ste. Marie, Ontario. To do this, Dr. Dave Roden of the Canadian Forest Service needs to obtain more infested branch samples from New York or Chicago in winter 2001. He has a permit for cuttings from NY. For the hybrid poplar work, scientists at the University of Wisconsin (Ken Raffa and Glen Stanosz) would like to participate.

Project Deliverables: Project results directly support evaluation of ALB threat to regions of the U.S., as well as provisions of information that can protect forest and nursery industries and improve restoration in urban areas.

For example, hundreds of thousands of hardwood nursery trees are produced each year in the US. Many of these trees have been selected for various traits including tree form, growth rate, foliage color, and insect resistance. If scientists can successfully identify trees that are resistant to ALB then there is a good chance that these traits can be used in various breeding programs. If ALB becomes established in natural and planted forests in the US, then developing tree lines that have high resistance to ALB would be of great value to the US forest and nursery industries.

Work plan Number FS 02.06 Goal: Regulatory

Project Title: Public Response to Possible Control Measures for the ALB and Development of Guidelines for Communication with Homeowners about Eradication and Control of ALB.

Principle Investigator(s): Susan C. Barro, USDA Forest Service, North Central Research Station, Evanston, IL, and Karen Vigmostad, Department of Resource Development, Michigan State University, East Lansing, MI

Project Objectives/Approach: The project objectives are as follows:

- Assess homeowner beliefs and attitudes about current control strategies for ALB,
- Evaluate homeowner beliefs and attitudes toward possible new control strategies that are currently under development,
- Summarize results of research with respect to what strategies are preferred or acceptable, and what aspects of each strategy are most critical to homeowners,

- Develop a publication to communicate with homeowners about the beetle and various control strategies,
- Develop guidelines for city officials and scientists to facilitate their communication with homeowners about ALB infestations and management strategies necessary for eradication and control.

Update: a) interim findings/activities –A draft manuscript to summarize results of the focus group study with Chicago area residents about their response to possible control strategies for ALB is being prepared. Two tools to facilitate communication with homeowners about ALB are being developed. A draft of the ALB brochure for homeowners will be sent out to scientists and homeowners for review within the next few months. A "tip sheet" for public officials to facilitate communication with homeowners is in the early draft stages.

Project Deliverable: Strengthened public understanding and support of eradication activities.

Work plan Number: FS02.07 Goal: Regulatory

Project Title: Assessing Tree Care Professionals' Awareness and Knowledge about the Asian Longhorned Beetle and its Detection.

Principle Investigator(s): Cem M. Basman, Department of Forestry, Southern Illinois University and Susan C. Barro, USDA Forest Service, North Central Research Station.

Project Objectives/Approach: The project objectives are as follows:

- Measure awareness levels of tree care professionals about the ALB outbreaks in the Chicago area, including quarantine areas and regulations for handling tree material,

- Assess the ability and knowledge levels of tree care professionals about the diagnostic signs of ALB infestation and preferred tree species,

- Determine what sources of ALB information have been used by tree care professionals and how effective/credible they believe these sources to be,

- Based on the above information, gather and develop suggestions for how to improve communication with tree care professionals in assisting the detection of ALB.

Update: interim findings/activities – Researchers are in the process of gathering background information and making decisions on the best approach for data collection.

Project Deliverables: Involvement of tree care professionals in improved detection of ALB.

Work plan Number: FS02.08 Goal: Control

Project Title: Life History Studies of the Asian Longhorned Beetle

Principal Investigator(s) name, location and affiliation:
Robert Haack, Forest Service, North Central Research Station, E. Lansing, MI; rhaack@fs.fed.us
Therese Poland, Forest Service, North Central Research Station, E. Lansing, MI; tpoland@fs.fed.us
Toby Petrice, Forest Service, North Central Research Station, E. Lansing, MI; tpetrice@fs.fed.us
Ruitong Gao, Chinese Academy of Forestry, Beijing, China; ruitong@mail.forestry.ac.cn
Deborah McCullough, Michigan State University, Entomology, E. Lansing, MI, mccullod@msue.msu.edu
Laura Lazarus, Michigan State University, Entomology, E. Lansing, MI, lazarusl@pilot.msu.edu

Project Objectives:
(Objective 1): To determine ALB within-tree distribution by tree species and seasonal development.
(Objective 2): To understand behavioral aspects of ALB host selection and oviposition.
(Objective 3): To determine the overwintering life stages of *A. glabripennis* along a North-South gradient in China and compare with overwintering stages found in the United States.

Project Update: ALB within-tree distribution was studied in Chicago by sampling infested trees as they were cut throughout 1999. In Chicago, scientists sampled primarily Norway maple, as well as box elder, silver maple, green ash, and American elm. Most sampled trees were cut during the dormant period but a few were sampled during summer. For the winter-collected host material, we found ALB eggs, all larval instars, and a few dead adults. Scientists found no pupae in winter, but others did. ALB life-stage distribution was similar among all tree species examined. Attack started in the crown of all trees examined. The fact that nearly all ALB life stages can overwinter explains why adults are found throughout summer and fall in the US.

In China in 2000, scientists recorded ALB within-tree distribution on more than 80 trees, including elms, poplars, and willows. These trees were part of a larger insecticide study in Gansu Province, China. The ALB within-tree distribution in China had aspects that were similar as well as different from the pattern seen in the US. Overall, for trees that had heavy sucker growth on the lower trunk (which was especially common in poplars) there was a tendency for attack to initiate at about the same time on the lower trunk as in the crown. Given that adults conduct maturation feeding on twigs, the presence of stem suckers apparently increases the likelihood of initial attack occurring lower on the trunk.

In November 2000, RA Haack was going to travel with RT Gao to two or three other provinces in central and southern China (Henan, Hubei, and Fujian). However, for some unknown reason the visa request for Dr. Haack was not approved and so the trip was indefinitely postponed. The goal of this study was to evaluate how the distribution of ALB life-stages changes in China along a north-south gradient. Results from this study would be valuable to the US in case ALB is found in locations to the south of the current infestations in New York and Chicago.

The attack pattern on any given tree is related to the tree's branching pattern. Because ALB adults feed on twigs throughout their adult life, oviposition will likely occur first in areas with nearby branches. When suckers occur on the lower trunk, oviposition will occur in this area. These findings should influence how inspectors examine trees, especially trees with suckers.

During 2002, scientists propose to travel to at least three sites along a North-South gradient in China where high populations of *A. glabripennis* and heavily attacked trees are available. During each trip (winter and summer), they will cut down and dissect several heavily attacked trees at each location. At least two tree species will be evaluated at each location. They will record all A.. *glabripennis* life stages found. These results will be compared to results from infestations in the US.

Project Deliverables: Information on seasonal development in various locations in China will allow US state and federal plant health specialists to better predict how ALB seasonal development will occur in different parts of the US. Although ALB is currently found only in Illinois and New York, there is a chance that ALB or other related species will be found in other US cities in the future. Knowing how seasonal development varies across China will allow US specialists to better time their survey and management efforts.

Work plan Number: FS02.09 **Goal: Control**

Project Title: Insecticide Control of Asian Longhorned Beetle

Principal Investigator(s) name, location and affiliation:
Robert Haack, Forest Service, North Central Research Station, E. Lansing, MI; rhaack@fs.fed.us
Therese Poland, Forest Service, North Central Research Station, E. Lansing, MI; tpoland@fs.fed.us
Toby Petrice, Forest Service, North Central Research Station, E. Lansing, MI; tpetrice@fs.fed.us
Ruitong Gao, Chinese Academy of Forestry, Beijing, China; ruitong@mail.forestry.ac.cn

Project Objectives/Approach: The main objective is to test the efficacy of various systemic insecticides to kill ALB larvae and adults. A secondary objective is to investigate the within-tree movement of systemic insecticides and dye.

Project Update: In 2000/2001, scientists evaluated three systemic insecticides in the US and China: imidacloprid, azadirachtin (the active ingredient in neem seed extract) and emamectin benzoate. In the US, imidacloprid was tested on both boxelder and silver maple; azadirachtin and emamectin benzoate were tested only in boxelder. ALB larvae were inserted in cut branch samples from azadirachtin-treated trees 4-weeks post-injection in the Ansonia quarantine lab, and the samples were dissected 8 weeks later. Similarly, ALB larvae were inserted in branch samples from the imidacloprid- and emamectin-treated trees 12-weeks post-injection, and the branches were dissected 4 weeks later. Overall, very low mortality occurred. Residue analyses, which are not yet complete, may help explain the low mortality levels. Scientists are currently rearing cottonwood borers (CWB) and ALB larvae on artificial diet that was treated with various concentrations of imidacloprid and azadirachtin. So far, after 8 weeks of feeding, CWB have shown dramatic reductions in feeding and weight loss and some mortality when concentrations of imidacloprid and azadirachtin are greater than 0.5 ppm. Similarly, reduced feeding and weight loss were seen for ALB after 2 weeks of feeding on treated diet. In China, we injected 24 heavily infested and 24 lightly infested elms, poplars, and willows. Trees were injected with imidacloprid, azadirachtin, or emamectin benzoate. Four weeks after injection, four mating pairs of ALB adults were caged on the lower trunk of each lightly infested tree. Four months after injection, we cut down and dissected half of the trees. Mortality levels varied by ALB life stage, tree species, and insecticide. Overall, moderate levels of mortality were found for all three insecticides tested. The highest mortality levels were seen in the poplar trees with 81% mortality of large larvae and 100% mortality of adults (that were still inside the trees) for imidacloprid, 47% mortality of pupae and 55% mortality of adults for azadirachtin, and 14% mortality of large larvae and 100% mortality of adults for emamectin benzoate. Residue analyses 4-weeks post-injection indicated imidacloprid levels of 0.7-2.6 ppm in leaves and 0.7-1.6 ppm in twigs, and azadirachtin levels of 2.3-33.6 in leaves and 0.8-2.8 in twigs. Residue analyses are not yet complete for emamectin benzoate. In both the US and China, dye was injected into trees and then the trees were felled and split. It was noted that dye travels closely along the walls of the ALB galleries, but not all branches received the dye.

Scientists propose evaluation of ALB mortality in the remaining treated trees in China in summer 2001 and complete the residue analyses. They will conduct further residue analyses of trees injected simultaneously with dye and insecticides (at various doses and numbers of injection points) to determine the optimal technique for uniform coverage. They also plan to evaluate thiachloprid, which is a new second-generation chloronicotynil insecticide, related to and possibly more effective than imidacloprid.

Researchers will coordinate needs for space and ALB larvae with USFS staff at the Ansonia quarantine lab. Some studies and residue analyses will occur at the Canadian Forest Service quarantine lab in Sault Ste. Marie, Ontario. They will coordinate research plans in China with USFS, SFA, and CAF staff. Trips to China are scheduled for May, July, and November 2001, with replicate studies in the spring and summer of 2002.

<u>Work plan Number: FS02.10</u> **Goal: Survey**

<u>Project Title</u>: Acoustical Signals of the Asian Longhorned Beetle

Principal Investigator(s) name, location and affiliation:
Robert Haack, Forest Service, North Central Research Station, E. Lansing, MI; rhaack@fs.fed.us
Therese Poland, Forest Service, North Central Research Station, E. Lansing, MI; tpoland@fs.fed.us
Toby Petrice, Forest Service, North Central Research Station, E. Lansing, MI; tpetrice@fs.fed.us
Cyrus Smith, Instrumentation & Controls Division, Oak Ridge National Laboratory, Oak Ridge, TN, smithcm2 @ornl.gov

Project Objectives/Approach:
The main objective is to identify unique acoustical signal descriptors associated with the presence of the ALB larvae in live trees and crating materials and to develop a prototype ALB acoustic detector.

Project Update:
Scientists conducted several studies in 2000/2001, both in the US and in China. In the US, we recorded the acoustic signals that are generated by wood-feeding insects as they tunnel in wood. They recorded ALB as well as several native wood borers (cottonwood borer, linden borer, locust borer, red oak borer, sugar maple borer, and white spotted sawyer). In China, they recorded from ALB-infested elms, poplars, and willows. Analyses of the acoustic signals were conducted by cooperators at the Oak Ridge National Laboratory. Analyses indicated that ALB feeding sounds have unique signal descriptors. Nevertheless, cerambycids larvae have many sounds in common as well. Recordings were made to compare different insect species, tree species, and distances from the sensor. Overall, it appears that a detector can be made that will discern ALB feeding noises from the feeding noises of other common wood borers. In addition, it also appears that a more generalized (less discerning) detector can be constructed that will detect a variety of insect feeding sounds.

Scientists will continue to record the sounds of other woodborers as we locate new species. In addition, they will do more studies to see how larval size affects feeding sounds, and how these sounds are modified by wood moisture content. They will also locate infested crating materials (or create it ourselves) and record and compare larval feeding sounds in live trees versus crating.

Researchers will work with ALB-infested materials in China and in quarantine labs in the US. We hope that we can record the sounds made by ALB larvae that are feeding in trees inside a quarantine greenhouse at Penn state University. Recordings from actual infested trees in Chicago or New York were made spring 2001.

Work proposed for FY 2002 includes further refinement of the ALB detection algorithm, evaluation of the system throughout the year and under different weather conditions, and miniaturizing the system. Studies will evaluate host material that is currently infested with ALB and other insects, including other wood borers and carpenter ants.

Scientists will evaluate variation in the sound pattern due to larval size, tree species, branch and tree diameter.

Project Deliverable: To efficiently and effectively detect ALB in trees and wood materials, using a portable acoustical detection device. The technology could reduce cutting of trees in urban eradication zones, based on readings that confirm which trees are ALB-free. Currently, all infested trees are discovered through visual inspection, including the use of tree climbers and bucket trucks. Advanced acoustic technology would greatly improve the chances for early discovery of infested trees. In addition, this acoustic technology could be used to locate trees infested with other exotic wood-boring insects if discovered elsewhere in the US.

Work plan Number: FS02.11 Goal: Regulatory

Project Title: Characterization of Asian Longhorned Beetle Populations and Biotypes.

Principle Investigator(s) name, location and affiliation: Melody Keena, USDA Forest Service, Northeastern Research Station, Hamden, CT

Project Objectives/Approach: Biological basis for estimating parameters for models of seasonal phenology, timing of exclusion and eradication methodologies; and assessing development and attack rates. Determine if specific biotypes of ALB are more successful at establishing from solid wood packing material under different climatic conditions.

Project Update:

Developmental Time-- Little if any larval growth occurs at 5° C and larval mortality increases at temperatures above 25° C. Work on the temperature thresholds for ALB development continues. Life table results continue to suggest that the New York and Illinois populations are different biologically. The life tables for the different populations will be redone using the new diet formulation this spring.

Attack Rates and Development in Cut Logs-- The results of cut log development studies on four maple species suggest that the female beetle is very discriminating when choosing oviposition sites that may partially explain why certain species are preferred over others. This also emphasized the fact that not all pits that are chewed will have eggs laid in them and that this varies between tree species and with host quality. The larval development results are consistent with what we have observed for cut infested wood and suggest that the beetle larvae are more likely to survive and develop from egg to adult in cut logs of softer wood maples than harder wood maples. Adults are more likely to emerge from infested wood that contains larger larvae that have entered the wood than logs that have small larvae or eggs just under the bark. This is particularly important since the softer wood maples are more likely to be used for firewood or packing materials such as crates.

More detailed work on the effects of chill on different stages will be done this year. Work on development will be compared between the artificial diet and tree hosts.

Project Deliverables: Biological information essential for improving timing of exclusion and eradication activities.

Work plan Number: FS02.12 **Goal: Other (Rearing)**

Project Title: Continuously Propagate in Quarantine ALB populations from Chicago, New York, and China

Principle Investigator(s) name, location and affiliation: Melody Keena, USDA Forest Service, Northeastern Research Station, Hamden, CT

Project Objectives/Approach: A consistently available, high quality supply of ALB for research purposes.

Project Update: Colonies from Ravenswood, Chicago, IL and Bayside, Maspeth, Sunnyside, and Flushing Meadows, New York, NY, are in their second or third laboratory generations in the Forest Service Quarantine Laboratory in Ansonia, CT. A new diet has been developed, based on the red oak borer diet that is easier to work with, has less contamination problems and shortens the rearing time for the beetle. Methods for rearing and handling each stage have been further refined.

The work of 10 Forest Service scientists and 7 collaborating scientists on ALB has been supported this last year. Dead adult and larval specimens for public education and outreach on ALB have been supplied to federal, state, university and public groups. There are currently ~1,600 large larvae being reared. Diet reared adults will be available from March through about August 2001. Costs of rearing one beetle have gone up due to changes in methods and price increases (salaries, utilities and supplies). Request provides for ongoing propagation of ALB in quarantine.

Current Cost of Rearing One Adult Beetle Using the Established Colonies

Item	ALB Stage	Supplies	Salaries	Quarantine	Overhead
Oviposition logs & twigs (1 wk)	Mating Adults	$ 5.74	$ 2.57	$ 0.49	$ 2.11
Chilling OV logs (1-3 months)	Eggs	$ 1.20	$ 3.10	$ 4.20	$ 2.04
Larval Rearing (16 weeks)	Larvae	$ 5.70	$12.92	$ 9.24	$ 6.69
Large Larval Chill (1-3 months)	Larvae	$ 0.52	$ 5.36	$ 4.20	$ 2.42
Larval Rearing to Pre-Pupae	Larvae	$ 2.00	$ 5.81	$ 1.96	$ 2.34
Pupil Handling	Pupae	$ 0.64	$ 2.39	$ 0.98	$ 0.96
Adult Setup and Feeding	Adults	$ 4.27	$ 1.65	$ 0.49	$ 1.54
TOTALS		$ 20.07	$33.80	$ 21.56	$ 18.11
		Grand Total		$ 93.54	

Project Deliverable: Supply of ALB for tests in quarantine related to dispersal and control.

Work plan Number: FS02.13 **Goal: Survey**

Project Title: Determine ALB Flight Propensity and Capacity in the Laboratory

Principle Investigator(s) name, location and affiliation: Melody Keena, USDA Forest Service, Northeastern Research Station, Hamden, CT

Project Objectives/Approach: Information on dispersal and capacity to spread that is fundamental to development of survey methods and protocols, and to determine establishment potential

Project Update: The results of flight trials suggest that about half of the newly emerged adults of both sexes will fly from a dry log, probably in search of food. Almost all beetles will walk up and/or down on the log before initiating flight. Well-fed females would be expected to stay on a host of good quality and chew oviposition pits rather than fly. Female flight would increase as host quality declines, either due to successive generations of use or high population numbers. When the host quality is poor >50% of females would be expected to fly within the first 30 minutes on that host and additional females would likely fly after they had time to chew pits to further assess the host quality. All males would be expected to fly at some stage in their life regardless of host quality since > 85% of all males flew in this study. Males may be more likely to stay on a log than fly when they have recently mated with a female. So, when population size is small males may fly more and females would likely fly less.

Project Deliverables: Observations made during the course of other studies were taken into account in planning the flight trials for this year. Future flight trials will build on the results already obtained. Study supports improved survey strategies and protocols.

Work plan Number: FS02.14 **Goal: Survey**

Project Title: Trapping system for Asian longhorn beetle and other Cerambycidae in China and North America

Principle Investigators: D.R. Miller, J.L. Hanula, G.L. DeBarr (USDA For. Serv. SRS) J. Sun, Z. Yang (Chinese Academy of Science)

Project Objective/Approach: Develop an efficient intercept trap for cerambycids in SE US; Characterize volatile profiles for tree species in se US; and test combinations of traps and lures in China.

Project Update:

Trap development – Tested 3 new designs along with the PheroTech multiple-funnel trap in 3 ecosystems in Florida and Georgia: (i) slash/longleaf pine; (ii) loblolly pine; and (iii) oak/white pine. Designs incorporated various intercept profiles (vane, pipe and toblerone) with bottom funnel of Lindgren trap. Ten replicates of each design were set in fall of 2000 and baited with high-release devices of ethanol and (-)-α-pinene (ca. 1 and 0.5 g/d, respectively). Significant numbers of 3 species of large Cerambycidae were caught in the slash/longleaf stands: Monochamus titillator, Arhopalus rusticus and Acanthocinus nodosus. Preference was found for the pipe design with the Lindgren trap ranking last. Beetles at the other sites were largely bark and ambrosia beetles.

Collapsible designs with the same profiles will be tested in spring and fall of 2001 in stands of loblolly pine and slash/longleaf pine. As well, we will test the effect of trap width, size of bottom funnel, and preservative vs. dichlorovos as a killing agent. Trials of best design will be conducted against PheroTech multiple-funnel trap, IPM Intercept trap and Hanula bole-mounted traps.

Volatile profiles – Host volatiles from boxelder (Manitoba maple) have been collected. The most abundant compound was diacetone alcohol. Characterizations of major pine and broadleaf species will be conducted in summer 2001 with GCMS analyses of Porapak collections.

Trials in China – Lindgren multiple-funnel traps baited with diacetone alcohol (ca. 0.5 g/d) were not attractive to ALB in China in late summer of 1999. The trial was repeated in early summer of 2000 with results yet to be received. Traps baited with ethanol (ca. 1 g/d) were weakly attractive to the cerambycids, Aroma moschata and Trichoferus campestris, and strongly attractive to the scarab, Potosia (Liocola) brevitarsis, but not attractive to ALB.
Results and trials coordinated with researchers in Canada (P. de Groot and R. MacInstosh) and representatives from Phero Tech and IPM Technologies.

Further trials proposed, to be conducted in China in 2002, after refinement of trap and stronger justifications for candidate compounds.

Work plan number: FS02.15 **Goal: Control**

Project Title: Microorganisms associated with Asian longhorned beetle

Principle Investigator(s): J.D. Podgwaite and V. D'Amico, USDA-FS, Hamden CT

Project Objectives: Discovery of microorganisms amenable to development as biopesticides

Project Update: Previously, scientists reported that bacteria representing several genera were isolated from the alimentary tracts of adult Asia longhorned beetles (ALB) collected in Chicago IL and Queens NY. The study was expanded to include various larval stages of ALB, mostly from logs from the Chicago infestations. Many of the bacteria found in the adults were also found in the larvae. The following also were identified: Leclercia adecarboxylata, Enterococcus casseliflavus, Curtobacterium flaccumfaciens, Rahnella aquatilis, Pseudomonas putida, Globicatella sanguinis, Klebsiella pnuemoniae ss rhinischeromatis, Serratia rubidae, Serratia ficaria, and members of the genus Staphylococcus. Some of the aforementioned are opportunistic pathogens in humans but as yet have not been tested for their pathogenicity for ALB.

Proposed for 2002: (1) determine pathogenicity of isolates for ALB, (2) continue search for pathogens in collaboration with colleagues at FS North Central Station laboratory, and (3) explore the symbiotic relationships of certain isolated bacteria with ALB.

Project Deliverable: New, biologically-based control tool for ALB.

Work plan Number: FS 02.16 Goal: Survey

Project Title: Location and probable function of ALB sensory receptors

Principle Investigator(s): Kathleen S. Shields, NRS

Project Objectives/Approach: Provide information on specific location and likely function of sensory receptors on antennae of male and female ALB that can be used to test chemical attractants.

Project Update: The surface morphology of the antennae of male and female ALB has been described and possible chemoreceptors have been located. There are no significant differences in surface structure of male and female antennae. Each antenna has at its terminus 11-20 basiconic pegs that are very likely to serve as contact chemoreceptors. In addition, segments 6-11 of each antenna have ~700 styloconic pegs that may serve as chemoreceptors; based on location and structure, their most likely function is close range chemoreception. Histological studies have identified some aspects of neural innervation. Work is ongoing to identify pore canals in these structures.

Results to date indicate that long range chemoreception is very unlikely.

Contact chemoreception is very likely and short range chemoreception is possible.

Continuing work will determine if the numerous styloconic pegs have pore canals, which would indicate that they function in olfaction.

Project Deliverable: Accelerate research to isolate chemical attractants that can be used in traps.

Work plan Number: FS02.17 Goal: Regulatory/Exclusion; and, Control

Project Title: DNA Analysis of ALB Populations

Principle Investigator(s): Jim Slavicek

Project Objectives/Approach: The development of nuclear DNA markers for use in determining the origin of ALB infestations, for identification of possible ALB samples, and to investigate ovipositing behavior.

Project Update: This project was initiated in FY 2001. ALB samples were collected in the Ravenswood area of Chicago. DNA isolation procedures are currently being developed for use on ALB larval samples. The process of developing DNA markers will begin once DNA isolation procedures are developed.

Continuation of study is proposed for FY 2002, including a collection trip to China.

Project Deliverable: Improved control methods and strategies through identification of ALB populations within and outside the U.S.

<u>Work plan Number:</u> FS02.18 **<u>Goal</u>: Control**

<u>Project Title</u>: Asian Longhorn Beetle Bacillus thuringiensis (Bt) Receptor

<u>Principle Investigator(s)</u>: Algimantas P. Valaitis

<u>Project Objectives/Approach</u>: To isolate and characterize the Bt receptor in ALB. This research will lead to development of in vitro assay for screening toxins to identify those that bind with high affinity, and hence may have potent insecticidal activity to ALB.

<u>Project Update</u>: This project was initiated in the Fall of 2000 and continued in 2001. A novel immunoaffinity protocol was developed for quickly extracting the Bt receptor from ALB larval gut membranes. Briefly, this involves using antibodies coupled to Sepharose beads for isolating the toxin-receptor complex from gut brush border membranes. The newly developed methodology will allow for rapid and efficient extraction of the Bt receptor from ALB. Several candidate coleopteran-active Bt toxins and the appropriate antibodies to generate the anti-toxin beads needed were procured. To optimize the conditions initial studies have been done with several lepidopteron and coleopteran insect species.

A key concern is to identify an effective Bt toxin against the ALB so that active toxin can be used to isolate and exploit the properties of the receptor for screening isolates of Bt. In discussion with Leah Bauer and other researchers we have initiated plans to coordinate our efforts to pursue bioassay studies at the quarantine facility in Ansonia ASAP.

<u>Project Deliverable.</u> Improved biologically-based control of ALB. For example, insecticidal proteins derived from Bt are the most extensively used biological insecticides. Despite the successful use of Bt in agriculture and forestry worldwide, little is known about pathways related to the insecticidal activity, and the development of resistance in target insects, especially in coleoptera. This work will contribute to ongoing cooperative research efforts aimed at understanding Bt toxin function and for development of new strategies for improved toxin usage.

<u>Work plan Number:</u> FS01.19 **<u>Goal</u>: Survey**

<u>Project Title</u>: Estimation of the dispersal rate of Asian longhorned beetle adults

<u>Principal Investigator</u>: David W. Williams, Northeastern Research Station

<u>Study Objectives/Approach</u>: To determine how far and how fast individual ALB adults may move as a means of estimating the potential rate of spread of infestations in the United States.

<u>Project Update</u>: FS scientist investigated adult dispersal at the Mt. Sorak National Park study site in South Korea using two approaches: capture-mark-recapture and harmonic radar. The first technique involved numbering individual beetles sequentially as they were encountered and noting their locations on individual trees at subsequent times

through daily censuses of the beetle population. Of the 29 adult beetles marked during the first week of fieldwork, 12 were recaptured 14-17 days after their initial capture. All but two beetles remained on the same tree or on neighboring trees (within a few meters) over the course of the observations. The two exceptions moved to trees about 100 m across an open space from the trees on which they were first captured. Of the same group of 29 beetles, five were not recaptured and five were recaptured only once during three weeks. It is not known whether they left the area or died unobserved.

FS scientists tested two types of harmonic radar tags in 2000. One was developed by USDA ARS in College Station, TX, and the other by Alberta Microelectronic Corporation in Canada. The new tags were light but durable and apparently did not hinder beetle flight. A new method was used for attaching tags to beetles, tying them dorsally across the pronotum and perpendicular to the long axis of the body with dental floss. As a result of the more durable tags and the improved method of attachment, tags stayed on the beetles for up to a week. However, the beetles were very rough on tags, and those that remained after a couple of days tended to become twisted and broken and to have a short detection range. Work to be continued in China in 2001 and is proposed for 2002.

Project Deliverable: Improved survey methods for ALB.

Work plan Number: FS02.20 Goal: Survey

Project Title: Natural resistance of forests to Asian longhorned beetle in East Asia

Principal Investigators: David W. Williams and Michael E. Montgomery, Northeastern Research Station

Study Objectives/Approach: To relate ALB population levels and damage to stand composition and other forest characteristics in closed forest stands in order to identify factors that make closed forests more or less susceptible to ALB.

Project Update: Scientists made significant progress toward achieving the objectives of this project by discovering native hosts of ALB in South Korea. Intensive searches at the site in Mt. Sorak National Park, another site 20 km away, and a third in Mt. Songni National Park in the center of the country turned up small populations of beetles on native Acer species. These limited results suggest strongly that ALB is endemic to South Korea. To date, the beetle has been found along the edges of forests, typically near roads. Searches deeper into the forest and away from roads have found few beetles, but these results are very preliminary and only suggestive.

In the coming season, we need to make an extensive survey of Acer forests across South Korea to locate study sites. Having found sites with appropriate habitats and beetles present, we will undertake intensive searches and map beetle populations, noting their host species and locations in the trees.

Project Deliverable: Information critical for projecting movement of ALB in the urban/wildland interface. As eradication boundaries expand in New York and Chicago, this information can help refine protocols for rapid detection in the urban/wildland interface and as ALB moves into urban forests.

Work plan Number: FS02.21 Goal: Control

Project Title: Use of Predaceous Nematodes for Control of Anoplophora glabripennis in Urban Trees in North America

Principal Investigator(s) name, location and affiliation: Leah Bauer, Forest Service, North Central Research Station, E. Lansing, MI
Deborah Miller, Forest Service, North Central Research Station,
E.Lee Solter, Illinois Natural History Survey, Champaigne, IL
James Cate, Integrated BioControl Systems, Inc., Aurora, IN

Project Objectives/Approach: Compare the efficacy of two native predaceous nematodes, Steinernema carpocapsae and Heterorhabditis marelatus, for control of Anoplophora glabripennis using two methods of delivery in urban trees.

Earlier research comparing four species of entomopathogenic nematodes, demonstrated the high virulence of Steinernema carpocapsae and Heterorhabditis marelatus against A. glabripennis in logs in the laboratory (Solter et al. in press). We plan to apply infective juveniles of these two species to infested trees in the field in China and North America according to methods tested previously in the laboratory. This methods involved applying sponges containing infective juveniles. In addition, we plan to evaluate the use of nematode-based sprays applied to tree trunks and branches. (Reference: Solter, L.F., Keena, M., Cate, J.R., McManus, M.L. and Hanks, L.M. 2001. Infectivity of four species of nematodes (Rhabditoidea: Steinernematidae, Heterorhabditidae) to the Asian longhorn beetle, Anoplophora glabripennis (Coleoptera: Cerambycidae). Biocontrol Sci. and Technol. (in press).)

Project Deliverables: Nematodes travel through galleries and are capable of finding their host. In addition, nematodes are capable of establishment within a tree, thereby, limiting the need repeated application, as required by conventional insecticides.

Work plan Number: FS02.22 Goal: Control

Project Title: Improved Delivery Methods for Systemic Insecticides to Control *Anoplophora glabripennis*

Principal Investigator(s) name, location and affiliation:
Robert Haack, Forest Service, North Central Research Station, E. Lansing, MI; rhaack@fs.fed.us
Therese Poland, Forest Service, North Central Research Station, E. Lansing, MI; tpoland@fs.fed.us

Toby Petrice, Forest Service, North Central Research Station, E. Lansing, MI; tpetrice@fs.fed.us
Ruitong Gao, Chinese Academy of Forestry, Beijing, China; ruitong@mail.forestry.ac.cn

Project Objectives/Approach: To develop improved guidelines and techniques for delivery of systemic insecticides in trees.

Scientists will evaluate several systemic insecticide delivery systems for their ability to ensure complete coverage within different species of trees and trees of different sizes. We will evaluate coverage within the cambial region where small larvae feed and deeper in the sapwood where larger larvae feed. We will test different dosages, injection devices, injection depths, and number of injection devices per tree. We will record the time that it takes for the injection process to be completed on each tree. We will evaluate these approaches using residue analyses of tree wood, twig, and leaf samples. Most of this project will be conducted in the US, but some parts of the study will be conducted in China so that currently infested trees in China can be compared against un-infested trees in the US. If permission is given to allow some infested trees to remain standing for a few weeks after discovery during the warmer parts of the year, we would prefer to test several such trees in the US.

Project Deliverable: Thousands of trees are now being injected each year in Chicago and New York using a standard approach. However detailed studies have not been conducted that evaluate within-tree coverage of the current technique to alternative techniques. The current technique often requires that individual trees be monitored for 4 hours while the insecticide is drawn into the tree. Another approach, which is called the “Systemic Tree Injection Tube” or STIT usually requires less than one hour per tree, and often less than 30 minutes/tree. Within-tree insecticidal coverage has not been evaluated between these two methods. If the STIT tube provides similar coverage to the standard technique now being used, then a considerable saving would be realized by switching the method of injection in the ALB eradication program.

Work plan Number FS02.23 **Goal: Regulatory**

Project Title: Evaluation of potential impacts of ALB on U.S. forests nationwide

Project Objectives/Approach: This proposal builds upon an assessment of the potential impact of the Asian longhorned beetle (ALB) in urban areas of the United States (Nowak et al., 2001). The project will analyze in FY 2002 the potential impact of ALB on U.S. forests nationwide. For each U.S. county, the total (and percent) number of trees and basal area that is susceptible ALB attack will be calculated. Forest data to determine susceptibility to ALB will be derived from the FIA national database. From these data, the following information will also be derived.

(a) Monetary value: Compensatory value of the forest will be calculated based on the Council of Tree and Landscape Appraisal (CTLA) guide methodology. To calculate the compensatory value, four variables are required:

- species (from FIA data set)
- diameter (from FIA data set)
- condition (average condition are to be derived from FHM data sets)
- location (i.e., forest)

Local species and diameter specific values used in the formula are derived from state value estimates (see Nowak et al., 2001 for more information on these estimates and CTLA formula). Compensatory value loss due to potential ALB impact will be calculated for each county.

(b) Range limitations: Average maximum and minimum temperatures, and annual precipitation data will be compiled for each county. These data will be used to help display potential range limitations to ALB infestation in the United States.

(c) GIS maps: Maps displaying the national ALB susceptibility data will be developed using Arc View. These GIS maps will be used to display, by county, U.S. forest information on:

- Proportion and total number of trees susceptible to ALB
- Proportion and total basal area susceptible to ALB
- Total compensatory value of trees susceptible to ALB
- Average maximum and minimum temperatures, and annual precipitation (these data can be used to develop potential range limitation maps for ALB)

Reference: Nowak, D.J., J. Pasek, R. Sequeira, D.E. Crane, and V. Mastro. 2001. Potential effect of *Anoplophora glabripennis* (Coleoptera: Cerambycidae) on urban trees in the United States. *J. Econon. Entomol*. 94(1): 16-22. (For copy of article, go to http://esa.edoc.com/server-java/Propub/esa/ec-v94n1.contents)

Project Deliverable: More precise projections of impacts to US forests from ALB will be determined. This information is important to setting ALB program priorities.

Work plan Number: FS02.24 **Goal: Regulatory**

Project Title: Windows Computer Program and Methodology to Assess the Potential Risk and Impacts of ALB in Urban Forests

Project Objective/Approach: The proposed study will develop a user-friendly Windows program that allows municipalities to assess the potential impact and risk of ALB to their urban forest resource. Using existing street tree inventory data, the program will determine:

- Potential number of trees at risk to ALB infestation
- Total leaf area at risk to ALB infestation
- Total potential risk to urban forest sustainability
- Total risk to forest functions and benefits

- Total tree biomass and carbon at risk to ALB infestation
- Total monetary value of the resource at risk to ALB infestation

The study will use: simple methods to assess the urban forest resource and user-friendly computer program to quantify the potential impact and risk to ALB infestation

Project Deliverables:

(a) An easy to use field manual and PDA (e.g. Palm Pilot) data recording program will be developed to allow cities to easily sample or inventory their urban forest resource. This type of data base is necessary to assess the potential risk to the forest from ALB attack. When gathering these data, data collectors will also be monitoring for evidence of existing ALB populations. Data from these assessments will be used for spreadsheets that serve as base data for the Windows ALB program.

(b) A user-friendly Windows desktop computer program will be developed to allow users to easily calculate the potential risk (and associated monetary value) of ALB infestations to their urban forest. Methods for field sampling and analysis will build off the existing Urban Forest Effects (UFORE) model methodology (Nowack and Crane, 2000) and ALB information (Nowak et al, 2001). This new computer program and users manual will allow local users a quick, easy and low-cost method of analyzing their urban forest for threat- thereby allowing development of appropriate management strategies to protect the urban forest resource from attack. Data will be displayed in both tabular and graphic format; program will allow for display of inventory data within GIS mapping programs.

Work plan Number: FS02.25 Goal: Control

Project Title: Develop Technologies for Direct Control of the Asian Longhorned Beetle Using Chemical Insecticides

Principal Investigator(s) name, location and affiliation:
Richard Reardon, Forest Service, Forest Health Technology Enterprise Team, Morgantown, WV rreardon@fs.fed.us

Other Cooperators:
Win McLane and Boade Wang, USDA-APHIS-PPQ, Otis Plant Protection Laboratory, Otis ANG Base, MA
Ruitong Gao, Chinese Academy of Forestry, Beijing, PRC

Project Objectives/Approach:
1) Determine the efficacy of systemic chemical insecticides applied as cover sprays and trunk and soil injections to adult and immature life stages of ALB using natural populations in China and; (2) determine residue profiles and environmental persistence of insecticides in soil and tree components (e.g. xylem, phloem).

Project Deliverable: Preventative and direct control tactics for controlling the spread and impact of ALB.

Updates: Preliminary data indicates the systemic insecticide, Merit, injected via Maugets provides the most efficacious systemic treatment. This effort has provided the much-needed data for the treatment of thousands of trees with systemic insecticides in the New York and Illinois eradication projects.

Work plan Number FS 02.26 **Goal: Control**

Project Title: Characterization of microorganisms associated with ALB

Principle Investigators: John Podgwaite, Vincent D'Amico and Roger Zerillo, Forest Service Research & Development, Northeastern Research Station. (email: Jpodgwaite@fs.fed.us)

Other Cooperators: The main focus of our microbial control effort is on bacteria and viral pathogens. Therefore, 1) microsporidia will be sent to Leah Bauer in North Central for further analysis and 2) fungi will be sent to Ann Hajek (Cornell University). James Slavicek, Northeastern Experiment Station, will be asked to perform DNA analyses on microorganisms that cannot be otherwise identified.

Project Objectives/Approach: Isolation and identification of microorganisms associated with the Asian Longhorned Beetle. These include (1) pathogens that may be further developed as microbial control agents and (2) symbionts that are required by Asian Longhorned Beetle for either food digestion or critical metabolic processes.

Project Deliverable: Study would be conducted from October 2001 to October 2003, with research in the U.S. and in China. Research would characterize the microbial ecology of the ALB; i.e., determine which microorganisms are often or always associated with the integument, alimentary system and other internal tissues and organ systems of larval and adult beetles. Research would identify these microorganisms to the closest possible taxonomic category using standard microbiological and DNA techniques and determine either their pathogenicity or symbiotic role for Asian Longhorned Beetle. Beetles studied will include healthy and naturally killed beetles from the US and China.

Updates: Isolation and identification of more than 20 species of bacteria and yeasts, at least two of which are likely to be symbionts. Further taxonomic work is necessary for the definitive classification of some biotypes and requires upgrading Microbial ID hardware and software. Bioassays are necessary to determine if any of the microorganisms are pathogenic to Asian Longhorned Beetles.

Work plan Number FS 02.27 **Goal: Control**

Project Title: Testing of Entomopathogenic Nematodes against the Asian Longhorn Beetle

Principal Investigators: Michael L. McManus, USDA/FS Northeastern Research Station and Leellen F. Solter, Illinois Natural History Survey

Other Cooperators: Dr. James Cate, Integrated BioControl Systems, Inc. will produce the nematodes needed for the studies. Dr. Melody Keena, USDA/FS will provide ALB larvae for laboratory testing. Dr. Lawrence Hanks will provide expert information about cerambycid biology.

Project Objectives/Approach: Research Timetable: September, 2001- December 2002 Evaluate and compare the efficacy of H. marelatus and S. carpocapsae against ALB. The bioassays for H. marelatus were preliminary and, although the data are encouraging, additional testing is needed to produce definitive information. First, we will determine the life stages of ALB against which the nematodes are active. For each life stage, we will determine the following characteristics of the nematodes: 1) the effective dosages for each nematode species, 2) time to ALB death, 3) numbers of infective juveniles produced in ALB cadavers, 4) in-lab host to host transmission.

Laboratory testing will take place at the Forest Service Quarantine Laboratory in Ansonia, Connecticut. Evaluation of nematode-killed larvae will take place at the Insect Pathology Laboratory of the Illinois Natural History Survey. On-log testing for the two isolates would be possible in Ansonia depending on availability of naturally infested logs or inoculated logs.

Deliverable: Biologically-based control option for ALB.

Updates: Solter et al. (in press) tested four species of nematodes against fourth and fifth instar ALB larvae. The ALB larvae were permissive to all four species, and infective juveniles were produced after host death. Two species, Steinernema carpocapsae and Heterorhabditis marelatus, produced particularly early host mortality and high numbers of juveniles; less mortality occurred with H. bacteriophora and H. indicus. Both S. carpocapsae and H. marelatus used in these experiments were isolated from soils in Indiana. Because only limited numbers of ALB larvae were available for testing, subsequent bioassays were conducted only on H. marelatus, which was selected because it is an actively searching species. Dosages of infective stage nematodes (LD50) were within the economically feasible levels expected for pests commonly treated with commercial products: 17 nematodes for early stage ALB larvae, and 349 nematodes for midstage larvae.

In addition to LD50 bioassays, preliminary tests were performed to determine if the infective nematodes could locate young ALB larvae within galleries in the cambium of cut logs. Nematodes on sponges (a commercial formulation) cut to approximately 2 cm2 were stapled to oviposition sites and moistened once during each of the first two days after their application. Larval mortality and the red coloration of cadavers produced by the nematode's symbiotic Photorhabditis sp. bacteria indicated that the nematodes were able to locate larvae within larval galleries up to 40 cm from the oviposition sites (Solter et al., in press). This project was funded by USDA/FS Cooperative Agreement #AG-00-CA-11242343-92 and the State

of Illinois, Dept. of Natural Resources.

References:
Solter, L.F., Keena, M., Cate, J.R., McManus, M.L. and Hanks, L.M. 2001. Infectivity of four species of nematodes (Rhabditoidea: Steinernematidae, Heterorhabditidae) to the Asian longhorn beetle, Anoplophora glabripennis (Coleoptera: Cerambycidae). Biocontrol Sci. and Technol. (in press).

USDA APHIS Methods Development Project Descriptions

Work plan Number: APHIS 02.01

Project title: Develop control technologies for the Asian longhorned beetle *Anoplophora glabripennis* Motschulsky in the People's Republic of China and in the United States

Principal Investigators & Affiliation: W. McLane, B. Wang, USDA-APHIS-PPQ, Otis Plant Protection Laboratory, Otis ANG Base, MA

Other Cooperators: Gao Ruitong, Chinese Academy of Forestry, Beijing, PRC; R. Reardon, USFS-SPF, Morgantown, WV

Project Objectives and Approach: (1) Evaluate and optimize the efficacy of systemic insecticides against Asian longhorned beetles (ALB) using natural populations; (2) determine the efficacy of insecticides used as cover sprays; (3) evaluate bait systems for control of ALB; (4) determine residue profiles and persistence of insecticides used against ALB. Field trials and laboratory tests will be conducted in China using naturally occurring ALB populations.

Project Deliverable: Effective means of controlling Asian longhorned beetle populations using insecticide-based technology

Updates: Results to date from this work have provided a large portion of the basis for the ongoing program of prophylactic treatment of thousands of trees with systemic insecticides in the New York and Illinois eradication programs.

Work plan Number: APHIS 02.02

Project title: Asian longhorned beetle treatment studies

Principal Investigators & Affiliation: W. McLane, D. Cowan, USDA-APHIS-PPQ, Otis Plant Protection Laboratory, Otis ANG Base, MA

Other Cooperators: USDA-APHIS Gulfport National Monitoring and Residue Analysis Laboratory, Gulfport, MS; J. Gittleman, USDA-APHIS-PPQ, ALB Eradication Project, Amityville, NY; D. Spilker, Bayer Corp.; J. Allen, Calvary Cemetery, Queens, NY; R.

Mungari, NY Dept. of Ag. and Markets, Albany, NY; A. Farran, J.J. Mauget Co., Arcadia, CA

Project Objectives and Approach: Overall objective is to determine optimal methodology for delivery of systemic insecticides against Asian longhorned beetle. Specific objectives are (1) determine the most effective method for delivering systemic insecticides; (2) define optimal parameters for soil injection as a method of pesticide delivery; (3) define optimal parameters for trunk injection as method of pesticide delivery. Field tests will be conducted in Calvary Cemetery in Queens, NY; trees treated with various application methods will be monitored by analyzing insecticide treatments in selected tissues at scheduled times following application.

Project Deliverable: Improved methodology and efficacy of insecticidal treatments for use in ALB eradication programs.

Updates: Results to date from this work have provided a large portion of the basis for the ongoing program of prophylactic treatment of thousands of trees with systemic insecticides in the New York and Illinois eradication programs.

Work plan Number: APHIS 02.03

Project title: Effect of tree size and application rate on Imidacloprid residues in treated trees

Principal Investigators & Affiliation: A. Sawyer, W. McLane, USDA-APHIS-PPQ, Otis Plant Protection Laboratory, Otis ANG Base, MA

Other Cooperators:

Project Objectives and Approach: The primary objective is to determine whether, and in what way, tree size and pesticide application rate affect residue levels of Imidicloprid in treated trees. Trees in different size classes will be treated with varying numbers of Maugets per inch of dbh, and selected tissues will be analyzed for Imidicloprid residues at selected times after treatment. A biologically based mathematical model will be used for analyzing the results.

Project Deliverable: Improved methodology and efficacy for systemic insecticide treatments agains Asian longhorned beetles

Updates: New work plan

Work plan Number: APHIS 02.04

Project title: Control technology for the Asian longhorned beetles: adult control using surface-applied baits and pathogens

Principal Investigators & Affiliation: D. Lance, V. Mastro, J. Francese, USDA-APHIS-PPQ, Otis Plant Protection Laboratory, Otis ANG Base, MA

Other Cooperators: A. Hajek, Cornell Univ., Ithaca, NY; Luo Youqing, Beijing Forestry Univ., Beijing, PRC

Project Objectives and Approach: (1) Develop delivery systems for topically applied pathogens and chemical insecticides for control of Asian longhorned beetles (ALB); (2) develop attract and kill technology using semiochemicals. Studies in China suggest that adult ALB spend a good portion of their time walking or resting on the larger stems of trees. Thus, spot or "banding" applications of insecticide or fungal pathogens, which have already demonstrated kill of beetles in tests by A. Hajek, could prove effective at eliminating adult beetles. In addition, semiochemical studies have identified several compounds that, while not functional as long-range attractants for enhancing trap catch, have shown possible short-range attraction that could prove useful in enhancing effectiveness of spot- or band-applied killing agents. A portion of the funds allocated to this project will support a cooperative agreement or contract for the production of fungal pathogens.

Project Deliverable: Control systems that could provide an effective additional tool for use in ALB eradication programs. These systems could provide a lower-cost and higher-application-speed alternative to systemic insecticides in areas at the perimeter of current program boundaries. Also, treatment of trees prior to removal could help avoid dispersal of beetles from falling trees into the surrounding area.

Updates: New work plan.

Work plan Number: APHIS 02.05

Project title: Translocation of systemic insecticides in forest and shade trees

Principal Investigators & Affiliation: V. Mastro, W. McLane, USDA-APHIS-PPQ, Otis Plant Protection Laboratory, Otis ANG Base, MA, R. Reardon, USFS-SPF, Morgantown, WV

Other Cooperators: To be determined

Project Objectives and Approach: Determine patterns of movement for systemic insecticides in forest and shade trees, and especially in preferred hosts of Asian longhorned beetles. Insecticides, dyes, and/or other traceable chemicals may be used.

Project Deliverable: Improvements in methodology and understanding of capabilities and limitations of systemic insecticides for control of Asian longhorned beetles.

Updates: New work plan; this will be handled through a cooperative or interagency agreement. Proposals are being solicited for this work.

Work plan Number: APHIS 02.06

Project title: Asian longhorned beetle: behavior, biology, control

Principal Investigators & Affiliation: B. Wang, D. Lance, V. Mastro, USDA-APHIS-PPQ, Otis Plant Protection Laboratory, Otis ANG Base, MA; J. Elkinton, Dept. of Entomology, Univ. of Mass, Amherst, MA.

Other Cooperators: Luo Youqing, Beijing Forestry University, Beijing, PRC; Gao Ruitong, Chinese Academy of Forestry, Beijing, PRC.

Project Objectives and Approach: Develop control and survey methodology for Asian longhorned beetle (ALB) by conducting cooperative studies in China and the United States. Funding for this project supports a cooperative agreement between the University of Massachusetts and APHIS-PPQ.

Project Deliverable: More effective tools for controlling and surveying for ALB.

Updates: Work conducted through this work plan has been an essential part of projects that have (1) demonstrated that chipping of infested trees is an effective method of killing immature ALB, (2) developed and evaluated systemic insecticides for ALB, which are now being used to treat thousands of trees in New York and Illinois and are considered a critical component of our eradication effort, and (3) led to the design of the current ALB survey protocols. In addition, services provided under this cooperative agreement have helped coordinate China-U.S. collaboration on commodity treatment projects and projects involving ARS and FS personnel.

Work plan Number: APHIS 02.07

Project title: Cooperative research on pathogens of longhorned beetles, *Anoplophora* species in China from 2001-2002 (He)

Principal Investigators & Affiliation: W. McLane, B. Wang, USDA-APHIS-PPQ, Otis Plant Protection Laboratory, Otis ANG Base, MA

Other Cooperators: He Xuexiang, Guangdong Forest Research Institute, Longdong, Guangzhou, PRC.

Project Objectives and Approach: (1) Conduct a field survey to catalog and determine the relative abundance of pathogens of *Anoplophora* beetles, and (2) evaluate virulence of pathogens to Asian longhorned beetles. Pathogens will be collected and, where appropriate, isolated, cultured, and bioassayed in the laboratory. Funds allocated to this work plan support a cooperative agreement between He Xuexiang, GDFI, and APHIS-PPQ.

Project Deliverable: More effective microbial agents for controlling *Anoplophora* beetles.

Updates: New work plan

Work plan Number: APHIS 02.08

Project title: Behavioral study and development of survey and management techniques for the Asian longhorned beetle, *Anoplophora glabripennis*

Principal Investigators & Affiliation: B. Wang, D. Lance, V. Mastro, J. Francese, USDA-APHIS-PPQ, Otis Plant Protection Laboratory, Otis ANG Base, MA

Other Cooperators: Luo Youqing, Xu Zhichun, Beijing Forestry University, Beijing, PRC; S. Teale, SUNY-ESF, Syracuse, NY; A. Zhang,

Project Objectives and Approach: (1) Evaluate possible semiochemicals identified in GC-EAD testing as antennally active as possible attractants of Asian longhorned beetles (ALB) bioassays in China; (2) characterize daily activity patterns of adult ALB; (3) determine factors influencing dispersal patterns of ALB; (4) identify cues used by ALB when orienting to host plants and other objects. These objectives will be met through a series of field tests and observations within natural populations of ALB in China. Funds allocated to this work plan support a cooperative agreement between Prof. Luo, BFU, and APHIS-PPQ.

Project Deliverables: (1) If proven biologically feasible, a trap or other sampling device that can be used in survey programs for ALB; (2) information useful in designing and optimizing survey and some control methods for ALB.

Updates: To date, compounds that are known to be antennally active have not proven to function as long-range attractants in field tests. However, a number of trap types have been evaluated and a functional trap type has been identified (but is not efficient enough in itself to be useful in survey programs). Patterns of activity of adult beetles have been characterized. Tests have demonstrated that beetles orient to vertical silhouettes, and additional tests are planned to examine effects of color, size and distance on their orientation.

Work plan Number: APHIS 02.09

Project title: Elucidation of pheromones in the Asian longhorned beetle, *Anoplophora glabripennis* by coupled GC-EAD

Principal Investigators & Affiliation: S. Teale, SUNY-ESF, Syracuse, NY; D. Lance, J. Francese, V. Mastro, USDA-APHIS-PPQ, Otis Plant Protection Laboratory, Otis ANG Base, MA

Other Cooperators: Luo Youqing, Beijing Forestry University, Beijing, PRC

Project Objectives and Approach: Identify attractants and other semiochemicals that are useful for survey or control of Asian longhorned beetles (ALB). Odors of beetles and host plants are collected on adsorbent material and subjected to gas chromatography and electroantennogram detector analysis (GC-EAD). This procedure identifies which chemicals in the odor mixture can be “smelled” by the beetles. Antennally active compounds are then identified by GC-mass spectroscopy, and, if appropriate, bioassayed to determine how they affect beetle behavior. Funds allocated to this work plan support a cooperative agreement between SUNY-ESF and APHIS-PPQ.

Project Deliverable: Attractants or other semiochemicals of ALB.

Updates: Over 20 antennally active compounds have been detected, and most of these have been identified. To date, none of the identified compounds have shown enough long-range attraction to be useful for baiting traps, but certain combinations appear to produce short-range attraction that may be useful for toxic baits or other similar purposes.

Work plan Number: APHIS 02.10

Project title: Spread models and survey design in new Asian longhorned beetle populations

Principal Investigators & Affiliation: A. Sawyer, V. Mastro, D. Lance, B. Wang, USDA-APHIS-PPQ, Otis Plant Protection Laboratory, Otis ANG Base, MA

Other Cooperators: J. Schaeffer, J. Gittleman, APHIS-PPQ-ER ALB Eradication Projects

Project Objectives and Approach: Develop mathematical models of spread and movement for ALB in newly infested urban, suburban, and forested areas. Models are based on analyses of extensive program survey, control, and host-tree data, along with data from dispersal studies that have been and are being conducted in China. Mathematical models will be used to generate optimal parameter values for survey efforts, and survey protocols will be designed accordingly.

Project Deliverable: Protocols and methodology for efficient and effective survey for new ALB populations, such as those found in the New York City and Chicago areas.

Updates: ALB eradication programs in New York and Illinois are currently using a 4-tiered system of survey protocols that are based closely on a prototype design that came out of this work plan. Those surveys have identified a number of newly infested sites in both program areas and are helping locate remaining pockets of ALB infestation, which is critical to the eradication effort.

Work plan Number: APHIS 02.11

Project title: Development and support of Quality Control practices for Asian longhorned beetle surveys

Principal Investigators & Affiliation: A. Sawyer, V. Mastro, USDA-APHIS-PPQ, Otis Plant Protection Laboratory, Otis ANG Base, MA

Other Cooperators: J. Schaeffer, J. Gittleman, C. Markham, APHIS-PPQ-ER; M. Stefan, APHIS-PPQ, Riverdale, MD

Project Objectives and Approach: Design and implement quality control measures (1) to assess the efficiency of visual survey programs at detecting Asian longhorned beetle (ALB) populations, and (2) to provide program managers with a tool to monitor the effectiveness of survey personnel. Simulated ALB damage and a variety of statistical procedures will be used.

Project Deliverable: (1) measures and assurance of the quality for ALB survey efforts, and (2) information of overall survey efficiency needed to accurately design, and allocate resources to, survey programs.

Updates: New work plan as of FY-2001; initial work is now in its formative stages.

Work plan Number: APHIS 02.12

Project title: Fumigation as a regulatory treatment for *Anoplophora* in Solid Wood Packing

Principal Investigators & Affiliation: A. Barak, R. Mack USDA-APHIS-PPQ, Otis Plant Protection Laboratory, Otis ANG Base, MA

Other Cooperators: Mr. Wang Yuejin, Chinese Entry-Exit Inspection and Quarantine (CIQ), Beijing; CIQ at Shanghai.

Project Objectives and Approach: Confirm that current APHIS T404-b-1, -2, -3 schedules for fumigating solid wood packing with methyl bromide and sulfuryl fluoride are adequate to kill *Anoplophora* larvae, pupae, and adults. New schedules will be developed if treatments in T404-b are shown to be inadequate. Tests will be conducted cooperatively between U.S. and Chinese scientists, using naturally and/or artificially infested wood samples. Residues will also be monitored to ensure that levels of fumigant are safe upon arrival of treated containers in the U.S.

Project Deliverable: Scientifically sound fumigation schedules for eliminating *Anoplophora* from solid wood packing materials.

Updates: Early studies under this work plan indicated that existing phosphine fumigation schedules were inadequate for disinfestations of solid wood packing materials. Validity of methyl bromide schedules have largely been confirmed, and work on sulfuryl fluoride is progressing. More low-temperature fumigation studies need to be done across the board.

Work plan Number: APHIS 02.13

Project title: Cooperative research on biology, ecology, and natural enemies of important species of Cerambycidae

Principal Investigators & Affiliation: B. Wang, V. Mastro, USDA-APHIS-PPQ, Otis Plant Protection Laboratory, Otis ANG Base, MA

Other Cooperators: Chinese Academy of Agricultural Sciences, Beijing.

Project Objectives and Approach: Review of available Chinese literature, as well as laboratory and field studies, will be used to document detailed information on the biology, ecology, and natural enemies of several important cerambycid insects. These insects include, in prioritized order, (1) *Chlorophorus annularis*, (2) *Purpuricenus temminckii*, (3) *Callidium villosulum*, (4) *Xylotrechus rusticus*, (6) *Tricoferus campestris*, 7) *Ceresium flavipe*.

Project Deliverable: Information that can be used in pest risk assessments that will provide a sound scientific basis for evaluating the quarantine status of these pests and help prioritize PPQ's detection survey efforts.

Updates: New project.

Work plan Number: APHIS 02.14

Project title: Development of treatment indicators and microwave technologies for the eradication of exotic pests in Solid Wood Packing Material (Penn State)

Principal Investigators & Affiliation: K. Hoover, M. Fleming, D. Agrawal, Penn State University, University Park, PA; V. Mastro, D. Lance, USDA-APHIS-PPQ, Otis Plant Protection Laboratory, Otis ANG Base, MA

Other Cooperators:

Project Objectives and Approach: The ability of microwaves to eliminate exotic insect pests from solid wood packing materials will be evaluated by treating wood samples in microwave ovens in both the U.S. and China. Temperature profiles will be developed based on power, time, and several variables. Kill of insects will be evaluated using both surrogate species and *Anoplophora* larvae. A variety of chemical groups, known to change physical properties upon heating, will be tested to determine their ability to remain on wood and change permanently when exposed to the appropriate range of temperature. Funding for this work plan supports a cooperative agreement between the Pennsylvania State University and APHIS-PPQ.

Project Deliverable: (1) Affordable technology that can be used by Chinese manufacturers to eliminate *Anoplophora* and other wood-boring insects from solid wood packing materials; (2) permanent markers capable of verifying that a sample of wood has received sufficient heat treatment to kill internal insects.

Updates: The ability of microwave radiation to kill insects in wood has been clearly demonstrated, and work to characterize effects of several variables (wood species, moisture content, wood thickness) on required time and power of treatment is progressing. A number of types of systems have been evaluated for potential as an indicators.

Work plan Number: APHIS 02.15

Project title: Cooperative research on host preference of *Anoplophora glabripennis* in China

Principal Investigators & Affiliation: B. Wang, V. Mastro, USDA-APHIS-PPQ, Otis Plant Protection Laboratory, Otis ANG Base, MA

Other Cooperators: Gao Ruitong, Chinese Academy of Forestry, Beijing, PRC

Project Objectives and Approach: Work will be conducted in China to assess the relative acceptability and suitability of selected Chinese and North American tree species as hosts for *Anoplophora glabripennis* (ALB). Choice and non-choice tests designs will be used in a series of common garden experiments that will extend over several years.

Project Deliverable: Reliable information on relative suitability of different host species for ALB. This information will be used to upgrade current pest risk assessments, prioritize our survey efforts for this insect, and in the design of control programs.

Updates: Saplings of 1 to 2 inches diameter have been planted in plots in Gansu Province, and tests are getting underway.

Work plan Number: APHIS 02.16

Project title: Host range of Asian longhorned beetle (*Anoplophora glabripennis*) among common urban landscape trees in North America

Principal Investigators & Affiliation: K. Hoover, J. Sellmer, S. Ludwig, Penn State Univ., University Park, PA; V. Mastro, D. Lance, B. Wang, USDA-APHIS-PPQ, Otis Plant Protection Laboratory, Otis ANG Base, MA

Other Cooperators:

Project Objectives and Approach: A wide variety of commonly planted landscape trees will be screened for susceptibility to Asian longhorned beetle (ALB). Tests will be run in quarantine greenhouses using potted trees. In addition, non-host (non-susceptible) species will be identified that can be used for re-planting in areas undergoing ALB eradication. Funding for this work plan supports a cooperative agreement between the Pennsylvania State University and APHIS-PPQ.

Project Deliverable: Reliable information on relative suitability of different host species for ALB. This information will be used to upgrade current pest risk assessments, prioritize our survey efforts for this insect, in the design of control programs, and in making decisions on which species to re-plant..

Updates: Testing is underway.

Work plan Number: APHIS 02.17

Project title: Development of artificial diets for Asian Longhorned Beetles

Principal Investigators & Affiliation: J. Tanner, USDA-APHIS-PPQ, Otis Plant Protection Laboratory, Otis ANG Base, MA

Other Cooperators: USDA-ARS, Philadelphia, PA

Project Objectives and Approach: Improved artificial diets and methods for producing diets will be investigated using twin-screw extruders and other alternative manufacturing methods. Diets will be evaluated for suitability for Asian longhorned beetle larvae in rearing evaluations. Funding for this work plan primarily supports an interagency agreement between the ARS and APHIS-PPQ.

Project Deliverable: Artificial diets that are easier to produce, last longer, and area adaptable to various containerization methods. This will improve survival and reduce handling of larvae, supplies used in rearing, diet change frequency, and rearing cost.

Updates: Initial tests of existing diet recipes, adapted to twin screw extruder use, have been conducted.

Work plan Number: APHIS 02.18

Project title: Improved methods and materials for rearing Asian longhorned beetles in the laboratory

Principal Investigators & Affiliation: D. Lance, J. Tanner, V. Mastro, B. Wang, USDA-APHIS-PPQ, Otis Plant Protection Laboratory, Otis ANG Base, MA

Other Cooperators: Pennsylvania State University, University of Illinois, SUNY-ESF, USDA-ARS, USDA-FS, and other cooperators who receive various life-stages of Asian longhorned beetles (ALB) for use in rearing programs, behavioral assays, diagnostic testing, or as identification aids.

Project Objectives and Approach: Evaluate novel approaches to rearing ALB in order to optimize (1) survival and reproductive potential of ALB in the rearing system and (2) cost-effectiveness of ALB rearing. Develop and assess the potential utility of new diets, novel rearing containers, adult artificial diets, artificial oviposition substrates, holding conditions, and other components of an insect rearing system.

Work under this plan also supports rearing of ALB for a variety of methods development projects both at Otis and among our cooperators.

Project Deliverable: A rearing system capable of efficiently producing insects for a variety of uses, including, for example, insecticide tests, behavioral assays, genetic control programs (such as sterile insect releases), etc.

Updates: Multi-generation rearing of ALB is being carried out successfully. An artificial diet has been developed and is being used routinely, but it needs improvements and alternative diets are under evaluation. *In vitro* hatching methods have been developed and are in routine use. A prototype artificial log has been developed. Live and dead insects have been provided to a number of cooperators and to in-house scientists for behavioral and insecticide-related testing.

Work plan Number: APHIS 02.19

Project title: Remote Sensing Technology for detection of Asian Longhorned Beetle Infestations

Principal Investigators & Affiliation: D. Lance, A. Sawyer, USDA-APHIS-PPQ, Otis Plant Protection Laboratory, Otis ANG Base, MA

Other Cooperators: to be determined

Project Objectives and Approach: Investigate the potential of remote sensing technology for identifying likely sites of Asian longhorned beetle (ALB) infestations. Obtain images from available libraries and analyze them to determine if there was a relationship between spectral plant stress indicators and sites of previous ALB finds. Contract to obtain new images using advanced imaging technology such as hyperspectral analysis and determine if trees or portions of trees with spectral characteristics indicative of stress will tend to harbor ALB infestations.

Project Deliverable: A method of identifying areas of plant stress similar to stress generated in response to ALB infestation, which could greatly increase the efficiency and cost-effectiveness of ALB survey programs, especially in areas beyond high-intensity surveys.

Updates: New project. Ideally this project should be picked up FS or ARS, as both have in-house capabilities in remote sensing. APHIS would be more than willing to work with either agency to determine the potential value of this technology.